Supelco®
Analytical Products

MERCK

Rapid and Robust Antibody Quantitation

Chromolith® WP 300 Protein A HPLC columns

Affinity chromatography is a selective technique which takes advantage of specific molecular interactions, for example antigen and antibody. The Chromolith® WP 300 Protein A HPLC column is designed to monitor monoclonal antibody titer and yield determination from cell-culture supernatants. This analytical scale procedure helps to optimize the titer of monoclonal antibody for the optimal time for harvest of the monoclonal antibody products.

Key Benefits:

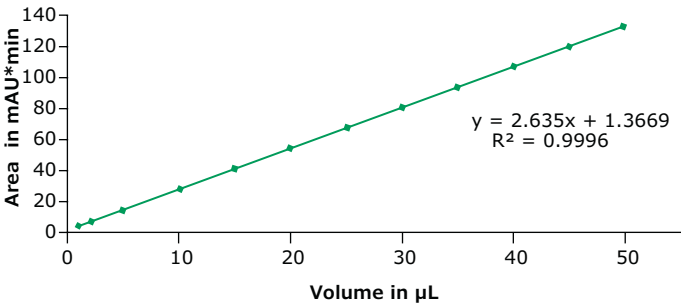
- Rapid and accurate titer determination across a wide range of injection volumes
- Consistent binding efficiencies even at high flow rates
- Expanded column lifetimes

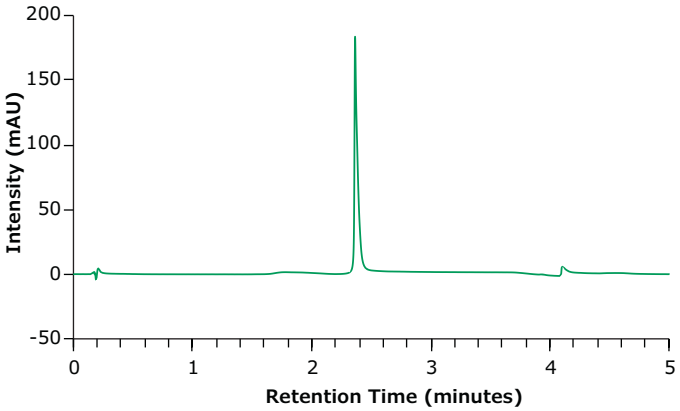
Demonstrated Linearity Across Injection Volumes

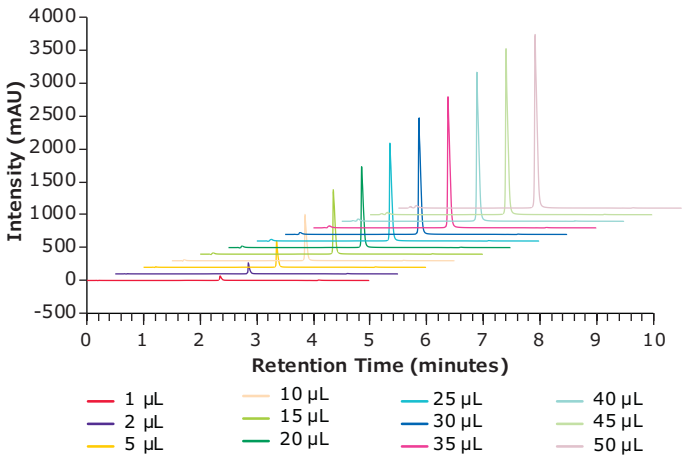
Columns:	Chromolith® WP 300 Protein A, 2.5 cm x 2.0 mm I.D.
Mobile phase:	[A] 100 mM Disodium hydrogen phosphate, pH 7.4; [B] 100 mM Disodium hydrogen phosphate, pH 2.5
Gradient:	

Time	%A	%B
0.00	100	0
0.50	100	0
0.55	0	100
2.60	0	100
2.65	100	0
5.00	100	0

Flow rate:	0.38 mL/min
Detection:	UV, 280 nm
Column temperature:	25 °C
Injection volume:	2.0 µL
Sample:	Gammanorm, 1 mg/mL, water





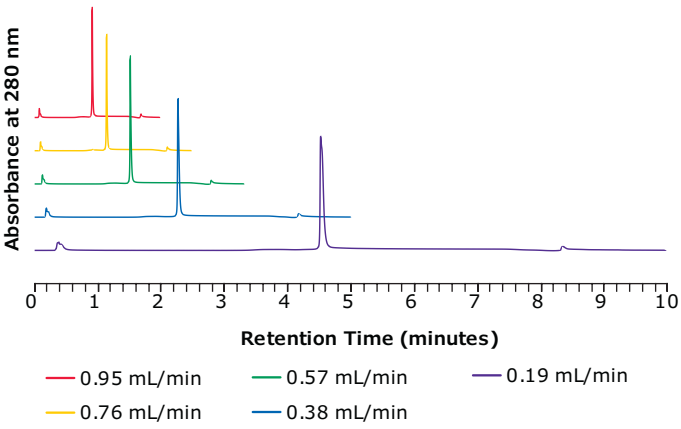


The life science business of Merck operates as MilliporeSigma in the U.S. and Canada.

Constant Binding Efficiency at Elevated Flow Rates

- High-speed separation at high flow rate due to excellent mass transfer properties of the monolithic skeleton
- At 1 mL/min on a 2 mm I.D. column, the total analysis time is less than 1 minute and the net column backpressure is only 24 bar
- Antibody binding is not affected by flow rate

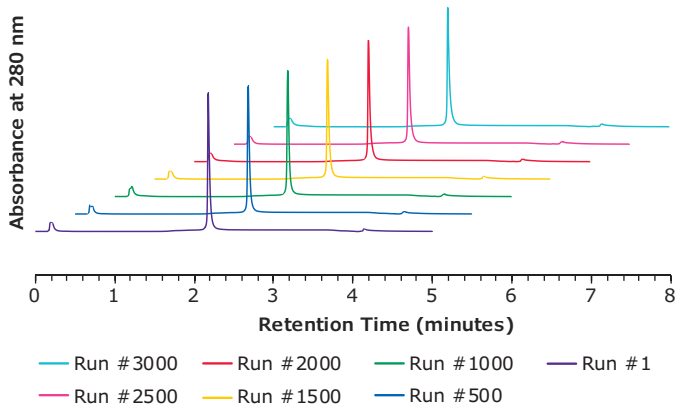
Flow Rate	Unbound area	IgG area	Pressure
0.19 mL/min	11%	89%	5 bar
0.38 mL/min	11%	89%	10 bar
0.57 mL/min	11%	89%	15 bar
0.76 mL/min	11%	89%	19 bar
0.95 mL/min	11%	89%	24 bar



Expanded Column Lifetimes

Chromolith® WP 300 Protein A column offers excellent reproducibility and stability for the long-term. In the below example, more than 3000 injections (> 80,000 column volumes, 6000 pH shifts) were performed on the same column, with no change in chromatographic performance.

Parameter	RSD
Retention time	< 0.3%
Peak Area	< 3.2%



Ordering Information

Part Number	Description	Length (mm)	I.D. (mm)
1.52358.0001	Chromolith WP 300 Protein A Column	25	2
1.52258.0001	Chromolith WP 300 Protein A Column	25	4.6

To place an order or receive technical assistance

Order/Customer Service: **SigmaAldrich.com/order**
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