

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.

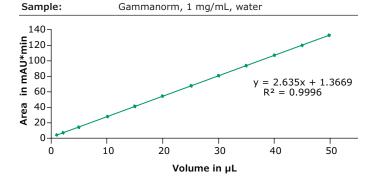
Demonstrated Linearity Across Injection Volumes

Columns:	Chromolith $^{\mbox{\tiny 8}}$ 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

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
	0.00 0.50 0.55 2.60 2.65	0.00 100 0.50 100 0.55 0 2.60 0 2.65 100

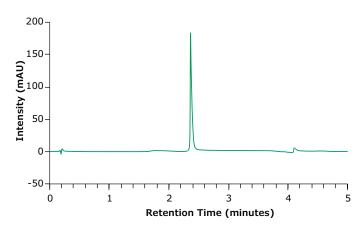
Flow rate:	0.38 mL/min
Detection:	UV, 280 nm
Column temperature:	25 °C

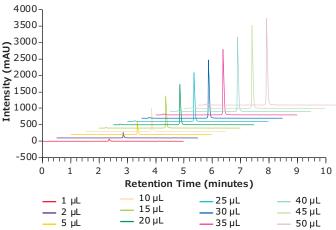
Injection volume:	2.0 μL



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



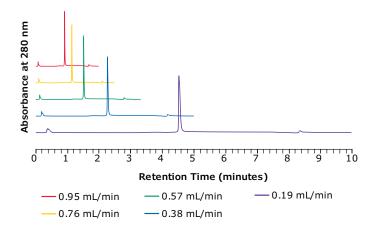




Constant Binding Efficiency at Elevated Flow Rates

- 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

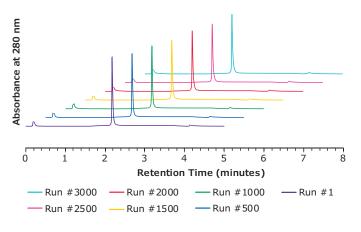
Flow Rate	Unbound area	IgG area	Pressure
0.19 mL/min	11%	89%	5 bar
0.38 mL/min	11%	89%	10 bar
0.57 mL/min	11%	89%	15 bar
0.76 mL/min	11%	89%	19 bar
0.95 mL/min	11%	89%	24 bar



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 Technical Service: SigmaAldrich.com/techservice

Safety-related Information: SigmaAldrich.com/safetycenter

MilliporeSigma 400 Summit Drive Burlington, MA 01803

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