

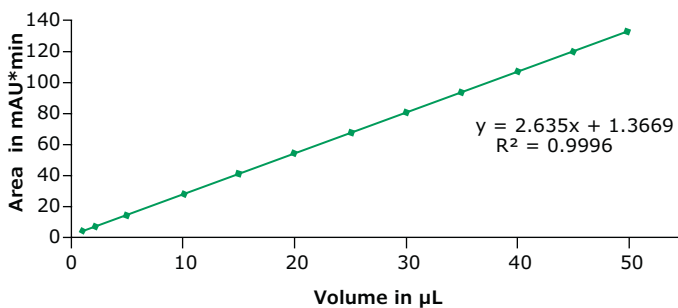
# Rapid and Robust Antibody Quantitation

## Chromolith<sup>®</sup> 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<sup>®</sup> 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.

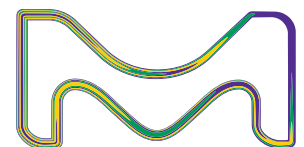
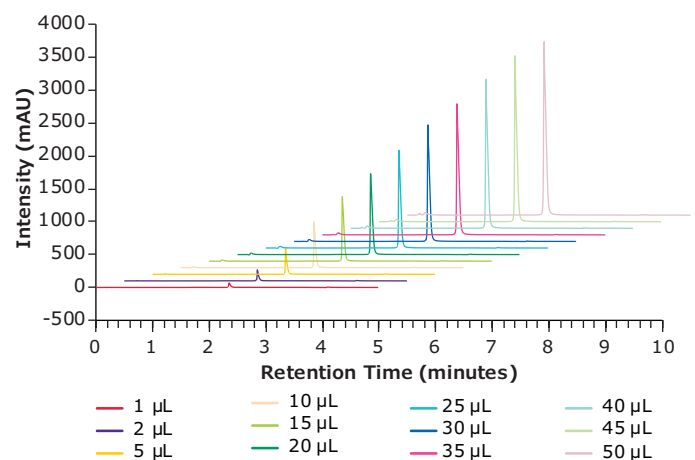
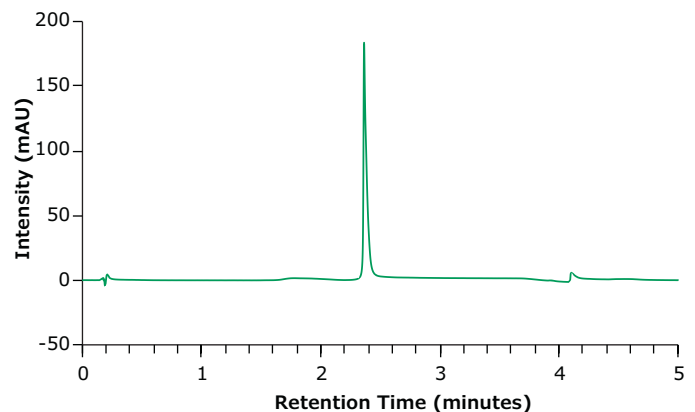
### Demonstrated Linearity Across Injection Volumes

<b>Columns:</b>	Chromolith <sup>®</sup> WP 300 Protein A, 2.5 cm x 2.0 mm I.D.		
<b>Mobile phase:</b>	[A] 100 mM Disodium hydrogen phosphate, pH 7.4; [B] 100 mM Disodium hydrogen phosphate, pH 2.5		
<b>Gradient:</b>	<b>Time</b>	<b>%A</b>	<b>%B</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
<b>Flow rate:</b>	0.38 mL/min		
<b>Detection:</b>	UV, 280 nm		
<b>Column temperature:</b>	25 °C		
<b>Injection volume:</b>	2.0 µL		
<b>Sample:</b>	Gammanorm, 1 mg/mL, water		



### Key Benefits:

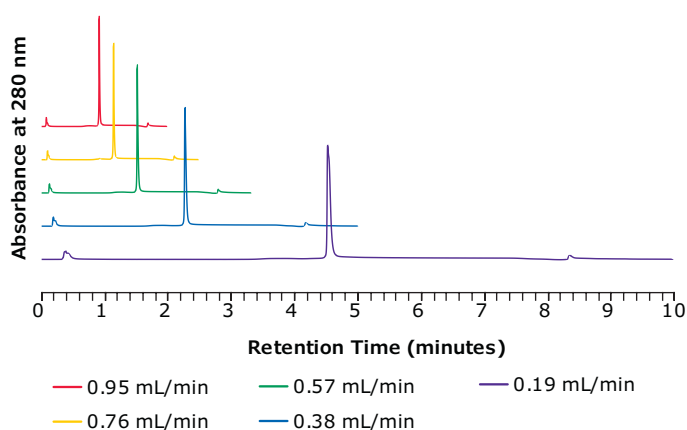
- Rapid and accurate titer determination across a wide range of injection volumes
- Consistent binding efficiencies even at high flow rates
- Expanded column lifetimes



## 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

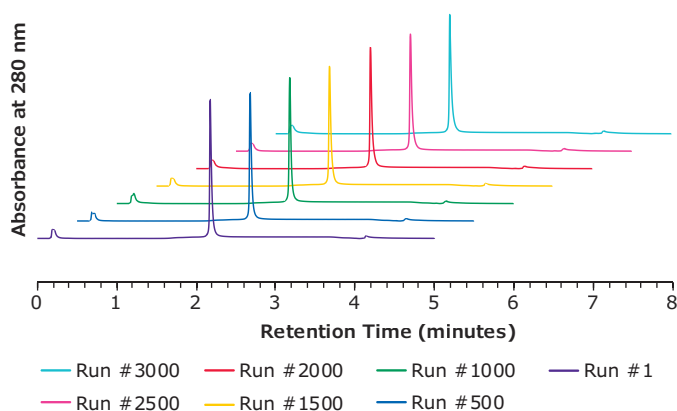
Flow Rate	Unbound area	IgG area	Pressure
0.19 mL/min	11%	89%	5 bar
0.38 mL/min	11%	89%	10 bar
0.57 mL/min	11%	89%	15 bar
0.76 mL/min	11%	89%	19 bar
0.95 mL/min	11%	89%	24 bar



## 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](https://www.sigmaaldrich.com/order)  
 Technical Service: [SigmaAldrich.com/techservice](https://www.sigmaaldrich.com/techservice)  
 Safety-related Information: [SigmaAldrich.com/safetycenter](https://www.sigmaaldrich.com/safetycenter)

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