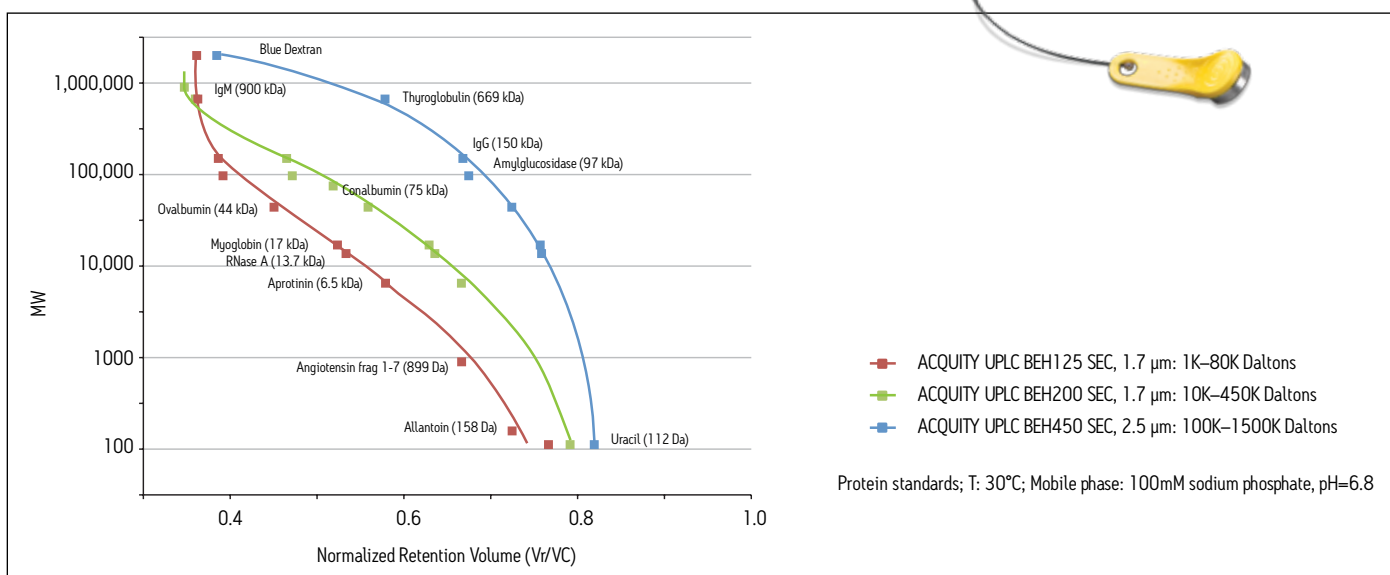


ACQUITY UPLC SEC System Solution

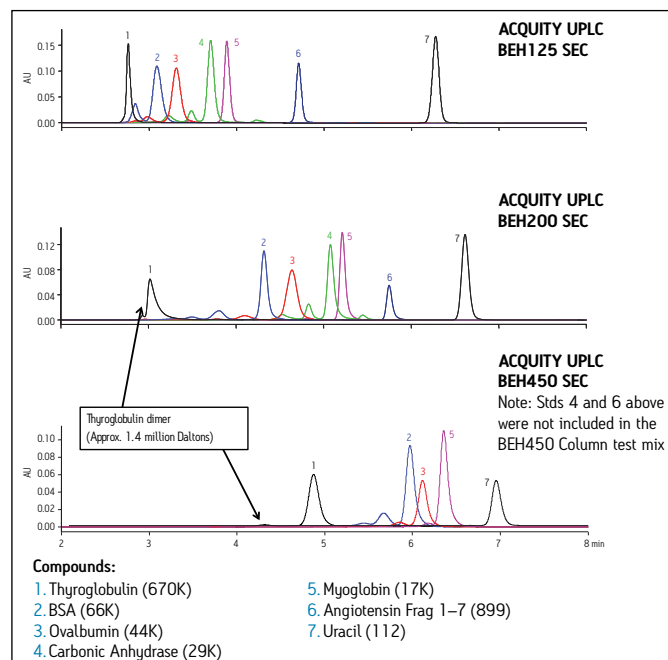
Waters ACQUITY UPLC offerings provide analysts with the enabling technology to see more and in less time. They have proven themselves to be valuable assets that improves the quality of collected data while increasing productivity. Biotherapeutic and biosimilar manufacturers can now choose the most appropriate UPLC-based, SEC column(s) (i.e., 125Å, 200Å, or 450Å pore size) to satisfy their application requirements based on this effective separation technology.



Calibration Curves on ACQUITY UPLC BEH125, BEH200 and BEH450 SEC Columns



Separation of Same Protein and Peptide Standards on ACQUITY UPLC BEH450, BEH200, and BEH125 SEC Columns

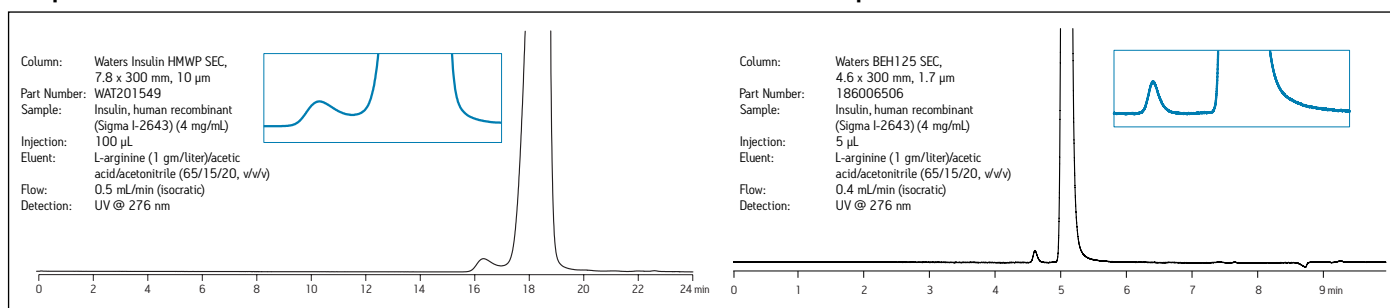


Columns	ACQUITY UPLC BEH125, 1.7 μm ACQUITY UPLC BEH200 1.7 μm	ACQUITY UPLC BEH450, 2.5 μm
Column Configuration	4.6 x 150 mm	
Mobile Phase	100 mM Sodium Phosphate Buffer, pH 6.8	
Weak Needle Wash	100% Milli-Q® Water	
Strong Needle Wash	100% Milli-Q Water	
Seal wash	90/10 water/methanol	
Samples: Diluted in mobile phase	Thyroglobulin 0.3 mg/mL BSA 0.3 mg/mL Ovalbumin 0.3 mg/mL Carbonic Anhydrase 0.3 mg/mL Myoglobin 0.3 mg/mL Angiotensin Frag. 1–7 0.1 mg/mL Uracil 0.1 mg/mL	Thyroglobulin 3 mg/mL BSA 5 mg/mL Ovalbumin 3 mg/mL Myoglobin 2 mg/mL Uracil 0.1 mg/mL
Injection Vol.	2 μL, Full Loop	
Flow Rate	0.3 mL/min	
Column Temp.	Ambient	
Detection Wavelength	UV @ 220 nm	UV @ 280 nm

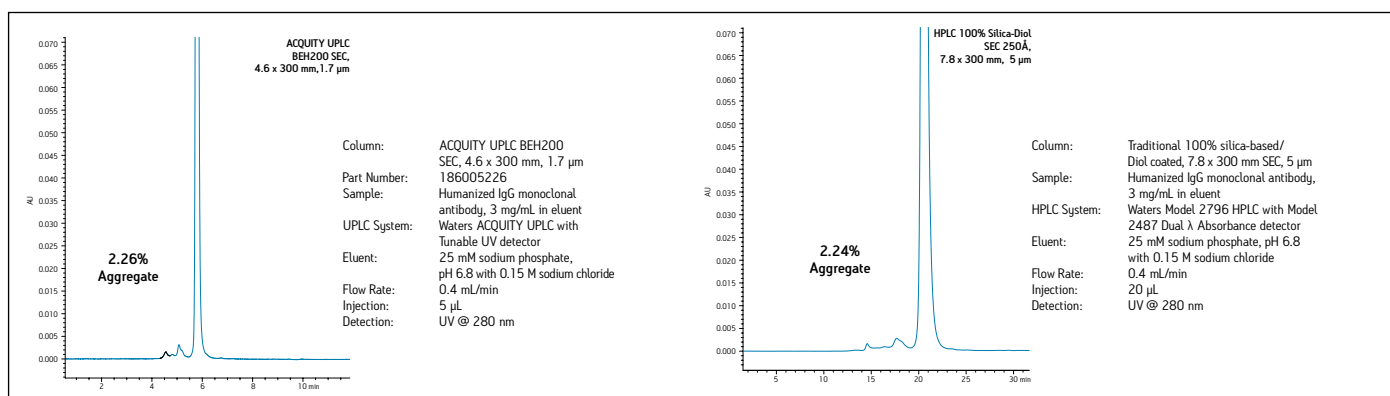
ACQUITY UPLC SEC Analysis of Therapeutic Insulin and Monoclonal Antibody

SEC is a well established and USP/EP approved method for the analysis of undesired, protein or peptide aggregates from active monomeric forms. As indicated in the figures below, Waters UPLC-based SEC separations deliver comparatively improved component resolution, while reducing analysis time and mobile-phase consumption. These attributes help get products to market faster with a consistency required by international drug regulatory agencies.

Comparative UPLC-Based SEC Benefits vs. Use of Traditional HPLC SEC for Biotherapeutic Insulin Characterization



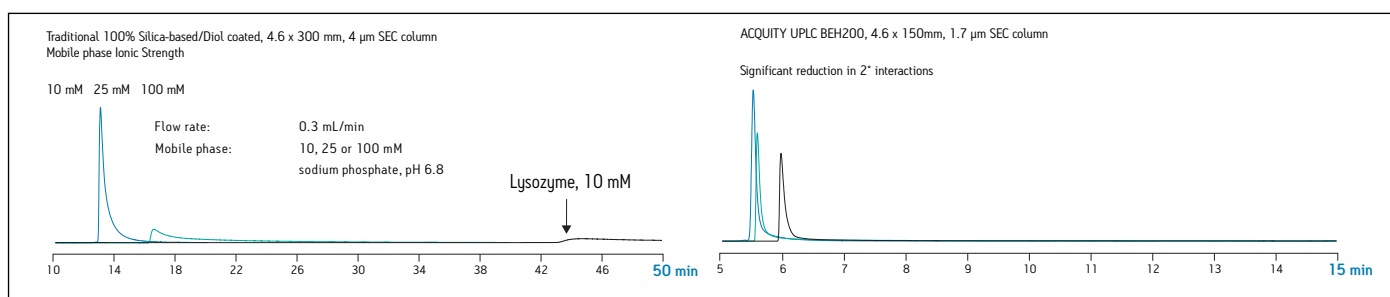
Comparative UPLC-Based SEC Benefits vs. Use of Traditional HPLC SEC for Biotherapeutic Monoclonal Antibody Characterization



Reduced Requirement for High Salt Concentration Mobile Phases

With conventional silica-based SEC column chemistries, undesirable secondary ionic interactions between the silica surface and basic proteins can result in long retention times and excessive peak tailing. Traditionally, the solution to this issue is the inclusion of high concentrations of a salt to compete for the charged sites on the surface of the silica. The unique surface chemistry of the ACQUITY UPLC BEH SEC particles significantly reduces these secondary interactions, resulting in the ability to use less aggressive mobile-phase salt concentrations.

Effect of Eluent Ionic Strength on the SEC Analysis of the Basic Protein Lysozyme on 100% Silica vs BEH SEC Particles



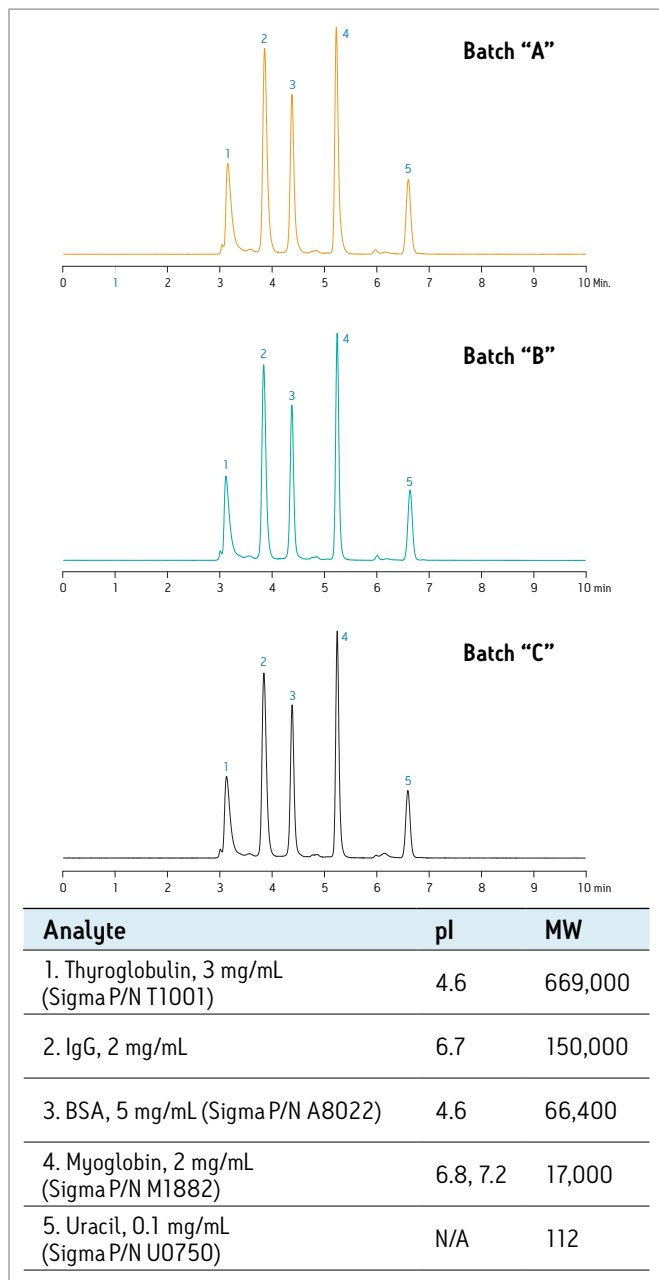
Stringent Manufacturing Quality Assurance Delivers Confidence in Results

All Waters ACQUITY UPLC column chemistries are synthesized from high quality raw materials in state-of-the-art ISO certified manufacturing facilities and are extensively QC tested throughout the synthetic process. In addition, each batch of ACQUITY UPLC BEH SEC material is specifically QC tested with relevant proteins and peptides to help ensure unmatched batch-to-batch consistency for supreme confidence in validated methods.

ORDERING INFORMATION

Description	Particle Size	Dimensions	Part No.
ACQUITY UPLC BEH125 SEC column	1.7 µm	4.6 x 150 mm	186006505
ACQUITY UPLC BEH125 SEC column	1.7 µm	4.6 x 300 mm	186006506
ACQUITY UPLC BEH125 SEC guard column kit	1.7 µm	4.6 x 30 mm	186006504
ACQUITY UPLC BEH200 SEC column	1.7 µm	4.6 x 150 mm	186005225
ACQUITY UPLC BEH200 SEC column	1.7 µm	4.6 x 300 mm	186005226
ACQUITY UPLC BEH200 SEC guard column kit	1.7 µm	4.6 x 30 mm	186005793
ACQUITY UPLC BEH450 SEC column	2.5 µm	4.6 x 150 mm	186006851
ACQUITY UPLC BEH450 SEC column	2.5 µm	4.6 x 300 mm	186006852
ACQUITY UPLC BEH450 SEC column w/ free standard	2.5 µm	4.6 x 150 mm	176002996
ACQUITY UPLC BEH450 SEC column w/ free standard	2.5 µm	4.6 x 300 mm	176002997
ACQUITY UPLC BEH450 SEC guard column kit	2.5 µm	4.6 x 30 mm	186006850
ELSD outlet tubing (0.004" id x 6" length)			430001562
0.005 x 1.75" SEC UPLC Connection Tubing, 2/pk			186006613
BEH125 SEC Protein Standard Mix			186006519
BEH200 SEC Protein Standard Mix			186006518
BEH450 SEC Protein Standard Mix			186006842

Notes: Size-exclusion chromatography may require modifications to an existing ACQUITY UPLC system. Please refer to "Size Exclusion and Ion-Exchange Chromatography of Proteins using the ACQUITY UPLC System", (715002147 Rev A) or "Size Exclusion and Ion-Exchange Chromatography of Proteins using the ACQUITY UPLC H-Class System", (715002909 Rev A) for specific recommendations.



Waters ISO 2001 Manufacturing and Testing Processes Help Ensure Outstanding ACQUITY UPLC BEH200 SEC, 1.7 µm Batch-to-Batch Reproducibility. This same approach is used to confirm consistent performance of the BEH125 and BEH450 SEC materials.

Waters

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