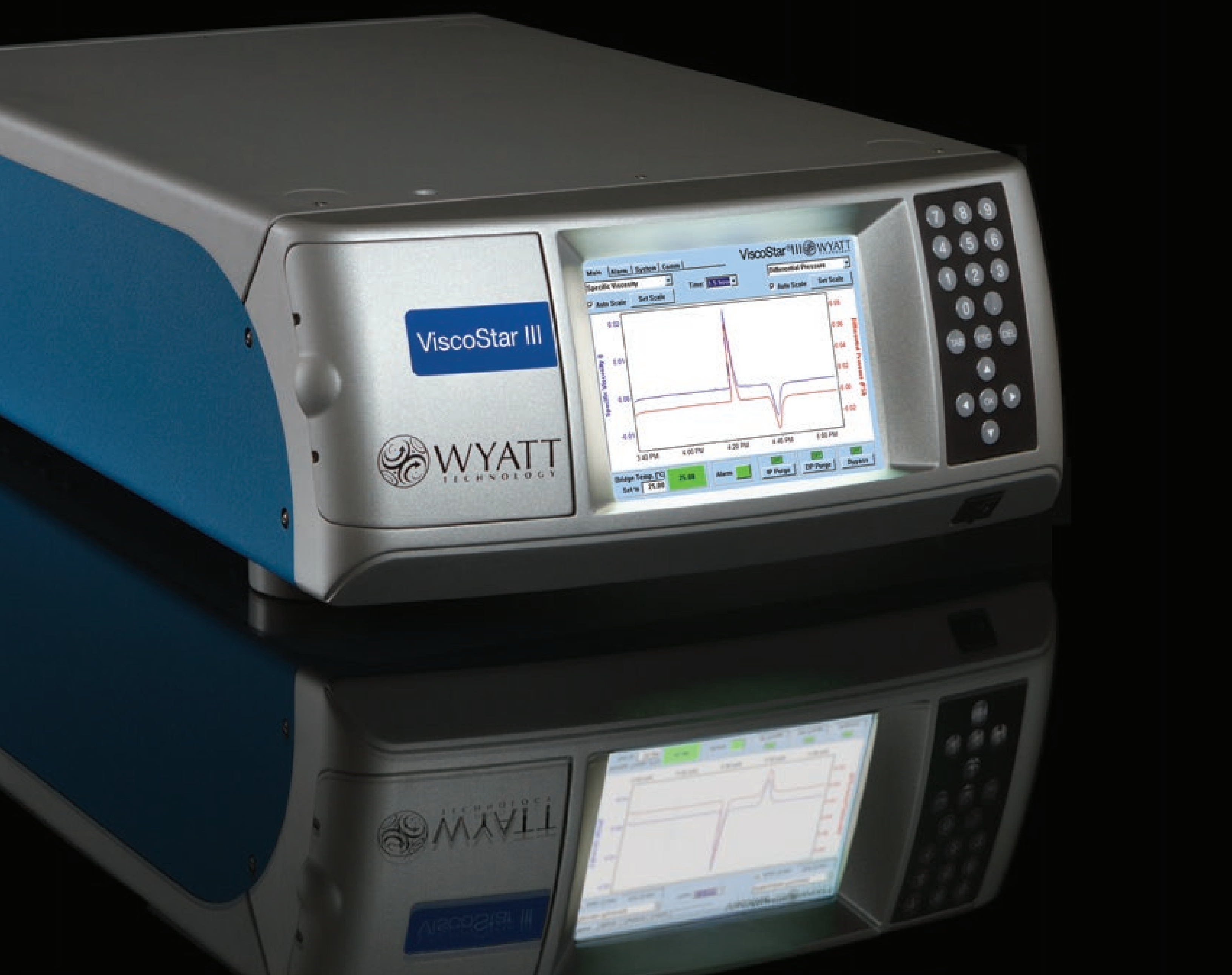
WYATT TECHNOLOGY CORPORATION

ViscoStar® III

Superior measurements of intrinsic viscosity
for macromolecular size and conformation



ViscoStar III

Characterize polymer branching and more with unmatched sensitivity

Benefits of SEC-IV

Intrinsic viscosity (IV) analysis is a versatile method for characterizing the physical properties of polymers and proteins. Combined with SEC (GPC) and a multi-angle light scattering (MALS) detector, the **ViscoStar III** determines:

- Intrinsic viscosity
- Conformation, branching ratios and drainage coefficients
- Mark-Houwink-Sakurada parameters relating the polymer's molar mass and intrinsic viscosity
- Hydrodynamic radii

Intrinsic viscosity is also useful for characterizing polymers that are *not* amenable to MALS analyses, such as:

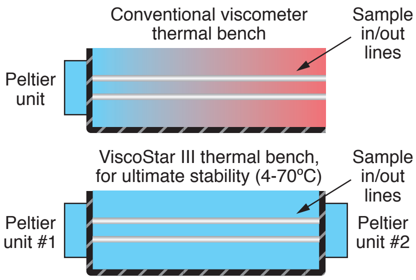
- Strongly fluorescent polymers
- Low *dn/dc* polymer/solvent systems that scatter light weakly

Wyatt's **ASTRA®** software package for SEC supports both Universal Calibration and Mark-Houwink-Sakurada analyses which utilize the ViscoStar *alone* to determine molar mass. With the highest sensitivity, fastest response and best thermal stability, the ViscoStar III is the premier differential viscometer for SEC.

Unique technology

Thermal stability

The ViscoStar III achieves highly uniform temperature *across the entire fluidics path* to ensure constant pressure differentials by means of an advanced thermal design, shown below.



A dual-Peltier design isolates the bridge from room-temperature variations.

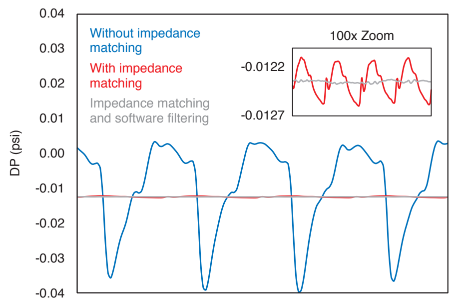
Reliability

Multiple safeguards have been incorporated in the ViscoStar III to enhance reliability and serviceability:

- Pressure transducers with all-316 stainless steel wetted surfaces for extended solvent compatibility
- System pressure sensor to detect clogs *and* prevent transducer damage
- Easy access to internal components

Pulse compensation

Most differential viscometers suppress pressure fluctuations electronically—at the cost of chromatographic resolution. The ViscoStar III introduces *complete* impedance balancing of the bridge that minimizes pump pulses *without* impacting resolution. A proprietary algorithm eliminates residual pulse amplitude.



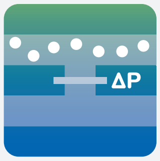
Patented pulse compensation technology eliminates pump fluctuations.

Thermal tuning

The resistive capillaries in a bridge must be finely balanced for optimal performance. Proprietary thermal bridge tuning ensures perfect balance in every run—with no moving parts.

Differential viscometry

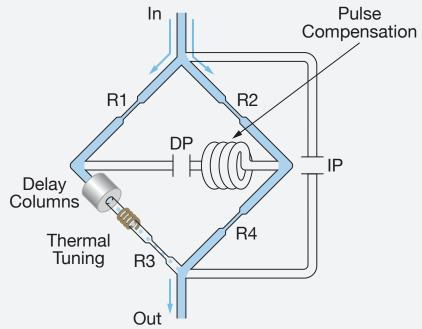
How it works



Differential viscometers measure very small changes in solution viscosity arising from the presence of macromolecules. This is accomplished with a balanced capillary bridge incorporating four capillaries of equal flow resistance R1-R4, shown below, and two pressure transducers IP and DP.

When pure solvent flows through all four capillaries, the bridge is balanced and DP reads zero. When sample is introduced, hold-up in the delay columns leads to an imbalance because sample flows through R1, R2 and R4 but solvent flows through R3.

The viscosity difference between sample and solvent creates a pressure differential on DP, which can be analyzed to determine the specific viscosity η_{sp}=4DP/(IP-2DP). The addition of an inline concentration detector (**Optilab® T-rEX™**) allows calculation of the sample's intrinsic viscosity.

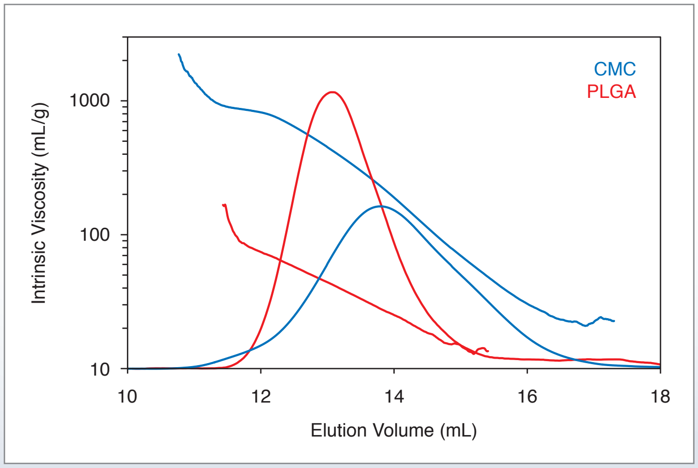


Without delay columns, the impedance of the capillary bridge would be fully balanced. The pulse compensation element matches the additional impedance of the delay columns, eliminating the effect of pump pulses on the DP transducer.

For more information

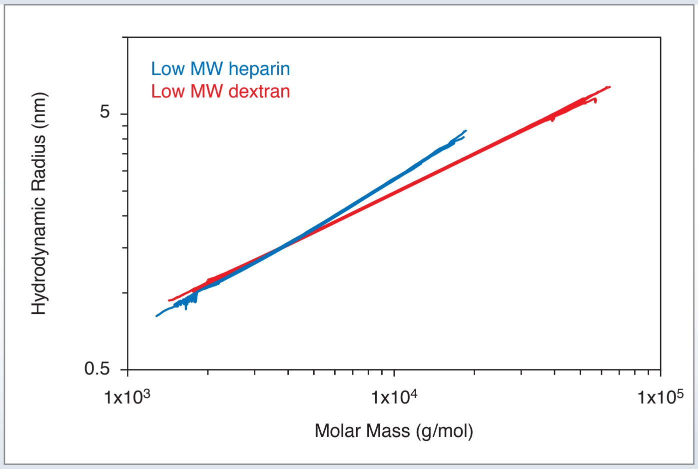
www.wyatt.com/SEC-IV
Intrinsic viscosity in conjunction with size exclusion chromatography

www.wyatt.com/ViscoStar
Additional ViscoStar III features



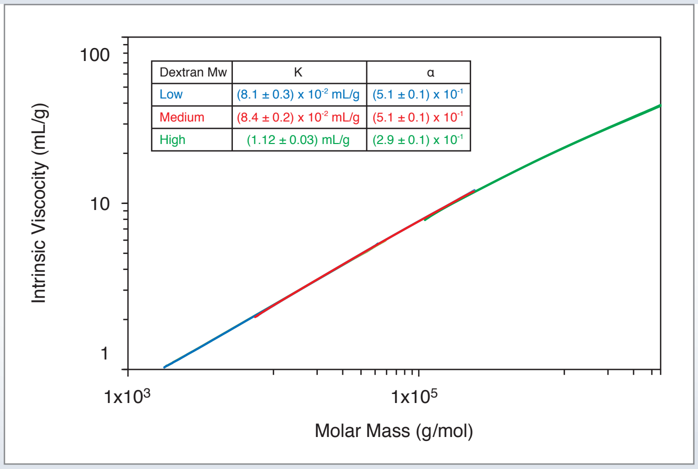
Intrinsic viscosity across a wide range

The ViscoStar III provides the highest available dynamic range and sensitivity in online viscometry. This graph shows the intrinsic viscosities of 1) poly(lactic co-glycolic acid), or PLGA, in THF and 2) carboxymethyl cellulose, or CMC, in aqueous mobile phase, both measured with a ViscoStar III and Optilab T-rEX, spanning a range from tens to thousands of mL/g. In other examples (shown below), the same instruments readily measure intrinsic viscosity values in the low single digits with excellent signal-to-noise, even at very low concentrations.



Small polymer sizing

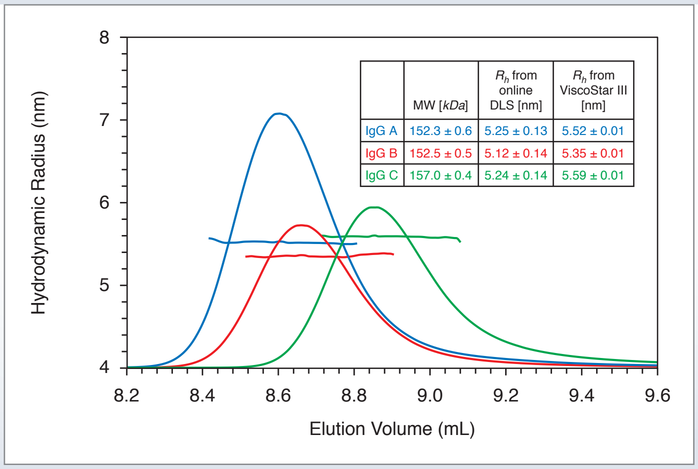
For a polydisperse polymer with an RMS radius less than 10 nm (or molar mass less than 100 kDa), viscometry is the best choice for measuring size and conformation. Here, the hydrodynamic radius of a low MW heparin and low MW dextran are plotted against molar mass determined with **DAWN® HELEOS®** MALS and Optilab T-rEX dRI detectors. Each sample was injected twice under four different SEC conditions, including different columns, mobile phases, and flow rates. The results demonstrate outstanding reproducibility and consistency of our triple SEC detection system.



Dextran Mw	K	α
Low	(8.1 ± 0.3) x 10^{-2} mL/g	(5.1 ± 0.1) x 10^{-1}
Medium	(8.4 ± 0.2) x 10^{-2} mL/g	(5.1 ± 0.1) x 10^{-1}
High	(1.12 ± 0.03) mL/g	(2.9 ± 0.1) x 10^{-1}

Conformational change with MW

A Mark-Houwink-Sakurada (MHS) plot shows intrinsic viscosity as a function of molar mass—revealing the polymer conformation. The MHS plots of low, medium, and high MW dextrans, shown here, indicate conformational change with increasing molar mass of the molecules. When a linear analogue is used as the reference, the ASTRA software further quantifies branching ratios and branch units-per-molecule.



	MW [*kDa*]	R_h from online DLS [nm]	R_h from ViscoStar III [nm]
IgG A	152.3 ± 0.6	5.25 ± 0.13	5.52 ± 0.01
IgG B	152.5 ± 0.5	5.12 ± 0.14	5.35 ± 0.01
IgG C	157.0 ± 0.4	5.24 ± 0.14	5.59 ± 0.01

Precise R_h measurement of proteins

Three IgG proteins with similar MWs eluted at different elution volumes. To understand their different elution properties, online DLS is typically used to measure the hydrodynamic radius, R_h, of a protein. However, online DLS *did not* provide reliable insights, in this case due to the relatively high experimental uncertainty of the DLS. The precise R_h values obtained with a MALS detector and the ViscoStar III, on the other hand, clearly revealed the causes. IgG B eluted later than IgG A due to its more compact structure; IgG C had a slight interaction with the SEC column.

Specifications

Differential viscosity	
Sensitivity (minimum detectable sample)	0.1 µg of 100 kDa polystyrene in THF at 1 mL/min, under typical GPC conditions
Differential pressure noise	< 0.05 Pa
Differential pressure range	6.9 kPa, protected against overpressure
Dynamic range	135,000:1
Linearity	0.5% of full scale
Differential pressure drift	< 2.5 Pa/hr
Capillary bridge autobalance	Thermal leak-free mechanism
Sample shear rate	5000 Hz with water @ 1 mL/min
Fluidics	
Maximum flow rate	Aqueous: 1.5 mL/min; THF: 3 mL/min
Delay volume options	16.2, 10.8 or 5.4 mL, with additional factory-installed option for 32.4 mL*
Maximum permissible pressure inlet to outlet	690 kPa
Wetted materials	316 stainless steel, PTFE, zirconium silicate, Kalrez®, Inconel, 26-1 Stainless Steel
Temperature control	
Range	4°C – 70°C
Stability	< 0.005°C
Ambient range	0°C – 40°C, 20% to 70% RH
Electronics	
Inputs/outputs	
Inputs	Autoinject contact closure, Alarm In, Analog In: 2 differential inputs, 16 bits resolution, range -10 V to +10 V
Outputs	Autoinject Retransmit, Alarm Out, Recycle Out
On-board computer	
Communications	Ethernet
Precision	20 bits
Integration time	0.1 sec to 20 sec, increments of 0.1 sec
Dimensions	60 cm (L) x 36 cm (W) x 17 cm (H)

*Volume of fluid through the ViscoStar until the retained sample elutes. Actual volume of fluid in the column is half this value.

Warranty: All Wyatt instruments are guaranteed against manufacturing defects for 1 year.

Wyatt Technology is committed to continual improvement. Specifications are subject to change without notice.

With installations in *more* than 65 countries, *more* than 12,000 refereed journal publications citing its instruments, and over 25 PhD scientists, Wyatt Technology is the **world's leading manufacturer of instruments and software** for absolute macromolecular and nanoparticle characterization. Our dedication to providing customers with comprehensive training and personal support make us the gold standard in this field.

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