Malvern

| Material relationships

MOLECULAR WEIGHT

MOLECULAR SIZE

MOLECULAR STRUCTURE

MALVERN
ADVANCED GPC/SEC
DETECTORS
SETTING THE STANDARD

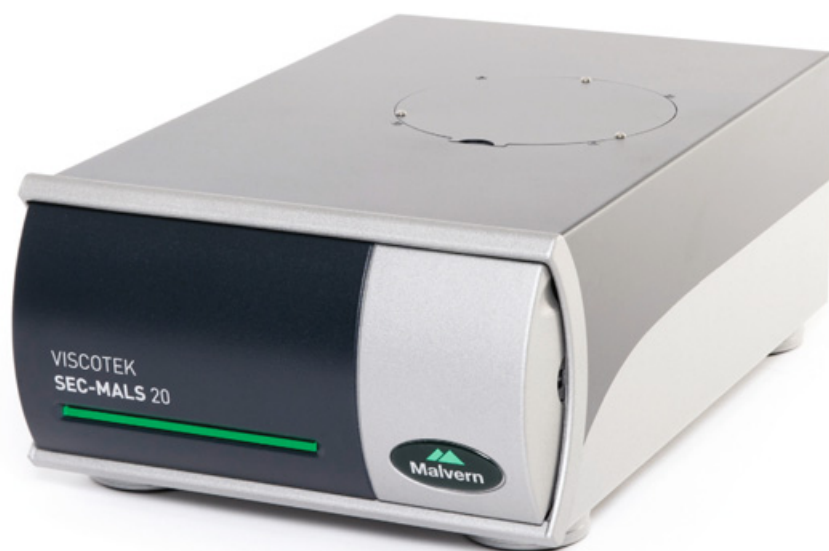# WHAT CAN ADVANCED GPC/SEC DETECTION OFFER YOU?

A modern research environment demands a complete understanding of a sample's molecular properties including accurate and reliable measurements of molecular weight.

Single detector GPC/SEC systems have been used for many years but cannot keep up with evermore demanding applications. The addition of advanced detection brings the greater understanding needed for today's samples.

- A **light scattering** detector: the heart of an advanced GPC/SEC system. Light scattering enables absolute molecular weight independent of structure and sample retention time

- A **viscometer**: probes the very structure of a molecule to give a sample's intrinsic viscosity. No other detector can match its sensitivity to structural changes such as branching

- A **refractive index** or **ultraviolet** detector: concentration measurement is the first step in any advanced GPC/SEC measurement and the Viscotek detectors' range includes various options.

The ultimate in advanced detection is the combination all of these detectors to get a complete understanding of the molecule being investigated.

Malvern's Advanced GPC/SEC Detectors

Through the innovative use of different measurement technologies, our detectors cover the full spectrum of GPC/SEC applications in the protein, polymer and polysaccharide characterization fields. The Malvern detector range can measure:

- **Absolute molecular weight, distribution and polydispersity**
- **Intrinsic viscosity and molecular structure**
- **Mark-Houwink parameters**
- **Molecular size (Rh & Rg)**
- **Branching number and frequency**
- **Composition**
- **2nd virial coefficient A_2 (B_{22}).**

Advanced GPC/SEC detectors can be used to upgrade existing GPC/SEC systems to more advanced setups offering the full range of applications but without the expense of a complete new system.

Choice and versatility drive our GPC/SEC range and our goal is to provide an advanced detection solution tailored to your specific needs.

All of the detectors in this brochure are available as individual modules to enhance your existing chromatography system, or as part of a complete GPC/SEC system. For more information about complete GPC/SEC systems, please refer to the separate brochure.

SYNTHETIC AND NATURAL POLYMER APPLICATIONS

The physical properties of a synthetic polymer like polystyrene, or a natural polymer like hyaluronic acid are strongly dependent on its molecular properties. Molecular weight, polydispersity, structure and (for copolymers) composition all contribute to determining the final product's properties and processability.

With more and more novel polymers entering the market, conventional measurements of molecular weight using a single detector are no longer sufficient. Our detectors make absolute measurements of these properties to cover a wide range of synthetic and natural polymer applications including:

- Polymer research
- Paints and coatings
- Food ingredients
- Drug excipients
- Tablet coatings
- Cosmetic and cosmeceuticals.

Molecular weight and molecular weight distribution can affect a polymer's **strength**, **durability**, **flexibility**, **toughness** and **degradation** rates. The different molecular weight moments also provide information on **flow properties** or **reactivity**.

Intrinsic viscosity is a direct measure of molecular structure and can be used to assess **branching**, which also affects **processability** and **flow viscosity**.

Copolymers combine the properties of their components. Measuring their **composition** will therefore help to understand the contribution of the individual components.

If you are interested in understanding these polymer properties better, take a look at the OMNISEC REVEAL or the Viscotek TDA.
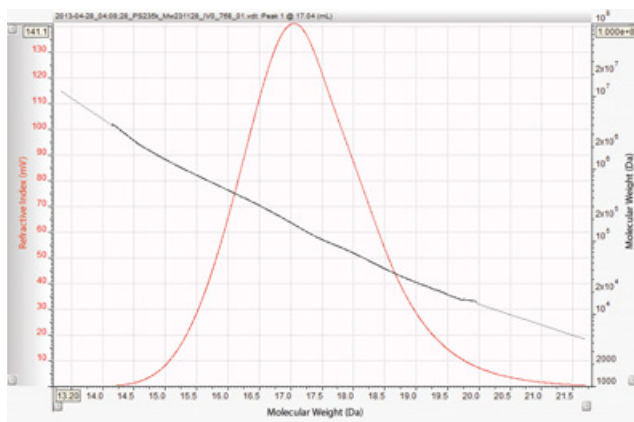

Absolute molecular weight measured by light scattering overlaid on the RI chromatogram
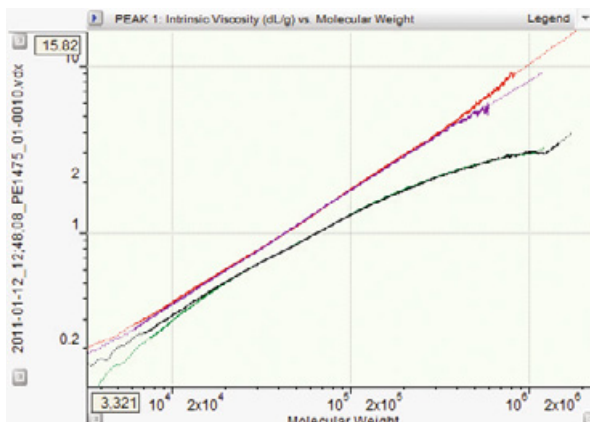
Mark-Houwink plot of molecular weight vs intrinsic viscosity for polymer structural elucidation.

PROTEIN APPLICATIONS

A protein's activity is strongly dependent on it being in the correct conformation and oligomeric state. Biopharmaceutical drugs must be aggregate-free and protein conjugates must be well controlled during research and as manufactured drugs. Single-detector GPC/SEC cannot meet the growing demands in this field but advanced GPC/SEC detection can help the protein scientist with all of these parameters, helping them to better understand the behavior of their protein of interest.
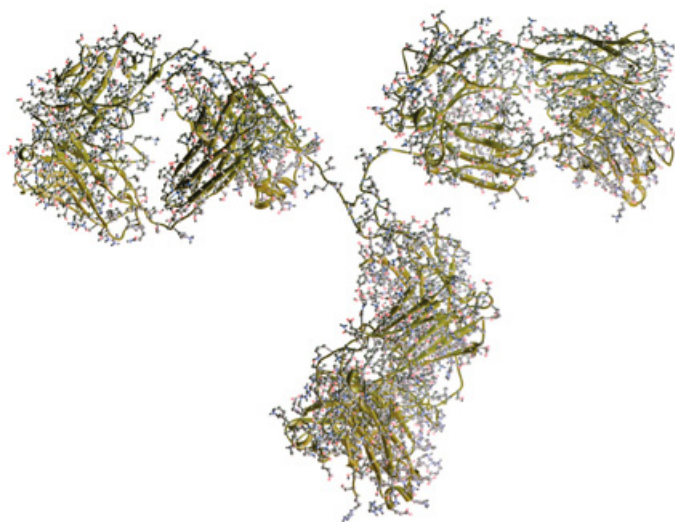
The molecular weight and size of a protein directly identifies its oligomeric state while polydispersity gives an indication of the purity of a sample peak.

Aggregates can be identified and their molecular weight, polydispersity and quantity all measured.

Conjugates such as PEGylated, glycosylated and membrane proteins can be characterized for their composition and molecular weight.

Intrinsic viscosity can be used to give an idea of **broad conformational changes** that relate, for example, to ligand binding.

If you are interested in understanding these protein properties better, take a look at the Viscotek SEC-MALS 20, OMNISEC REVEAL or the Zetasizer μV.
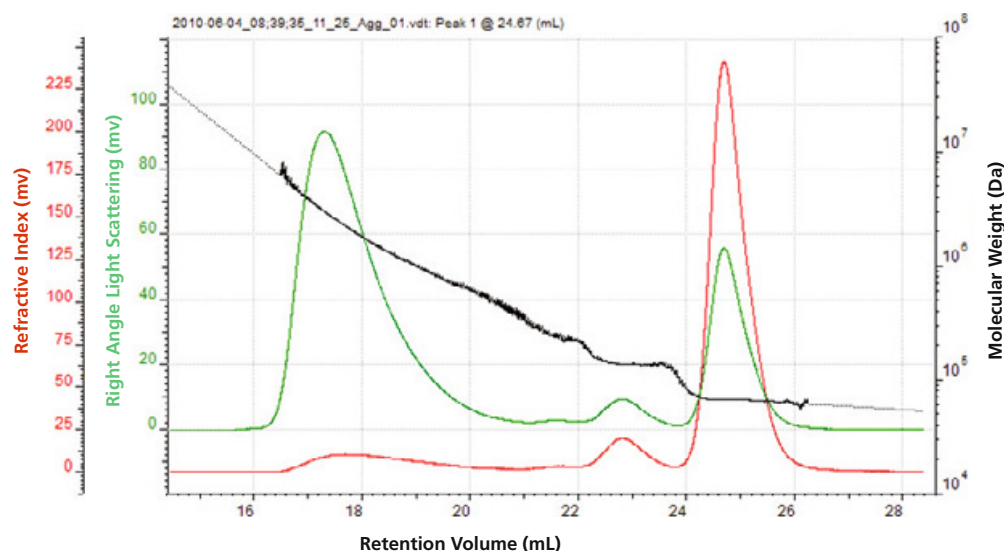
Protein oligomers and aggregates shown by Right Angle Light Scattering (RALS) (green) and Refractive Index (RI) (red) chromatograms overlaid with the absolute molecular weight (black).

OMNISEC **REVEAL**

Absolute Molecular Weight, Intrinsic Viscosity, Molecular Size, Concentration

OMNISEC REVEAL is Malvern's latest and most advanced integrated multi-detector system for superior GPC/SEC analysis. It includes refractive index (RI), UV/Vis absorbance, light scattering and intrinsic viscosity detectors for the characterization of synthetic polymers, natural polymers and polysaccharides, and proteins.

In combination, these detectors can measure:

- Sample concentration
- Absolute molecular weight and molecular weight distribution
- Polydispersity
- Intrinsic viscosity
- Molecular size (Rh & Rg)
- Molecular structure
- Mark-Houwink parameters
- Refractive Index increment, dn/dc.

An integrated design keeps all of the detectors in one compartment, affording multiple advantages:

- Inter-detector tubing is minimized reducing band broadening to improve data quality and result accuracy
- The detectors and the inter-detector tubing are all maintained at the same temperature to further improve data quality by maintaining baseline stability.

The result is unparalleled multi-detector data quality.

# OMNISEC **REVEAL** TECHNOLOGY

Absolute Molecular Weight, Intrinsic Viscosity, Molecular Size, Concentration

OMNISEC REVEAL's **refractive index detector** measures sample concentration. Its robust flow cell is kept in series with the other detectors for maximum sensitivity.

The **UV/Vis photodiode array** (200-900 nm) opens up absorbance measurements to a wider application range.

The **light scattering detector** combines the sensitivity of 90° Right Angle Light Scattering (RALS) with the accuracy of 7° Low-Angle Light Scattering (LALS). Its superior sensitivity makes it ideal for measuring low molecular weight, low concentration or low dn/dc samples, while its 18 μL flow cell minimizes band broadening.

OMNISEC REVEAL's **digital differential viscometer** includes a unique self-balancing bridge which can be easily replaced by the user. Its new 316 stainless steel pressure transducers improve baseline stability, sensitivity and robustness and means few limitations with salts or pH.

Intrinsic viscosity
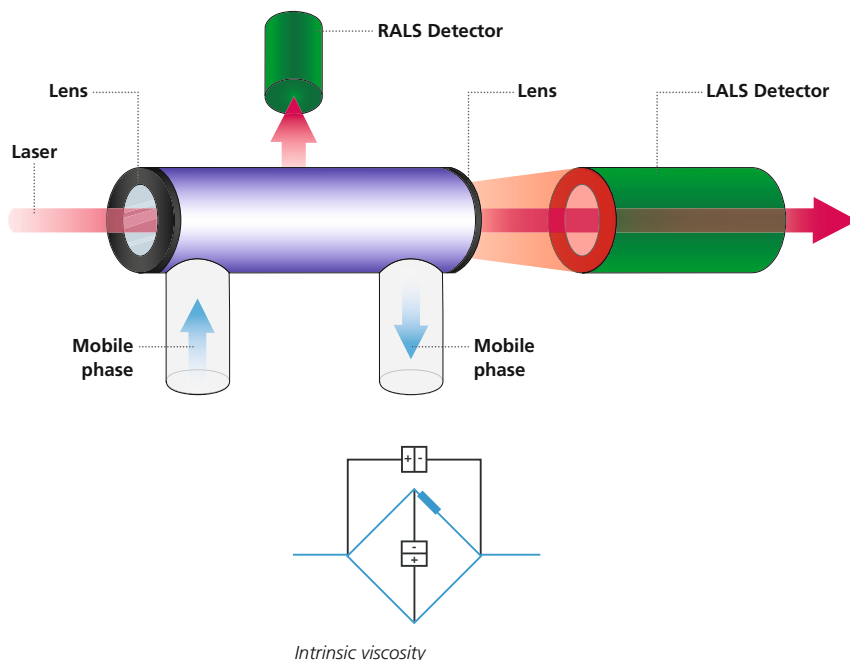
OMNISEC SOFTWARE v10

Absolute Molecular Weight, Intrinsic Viscosity, Molecular Size
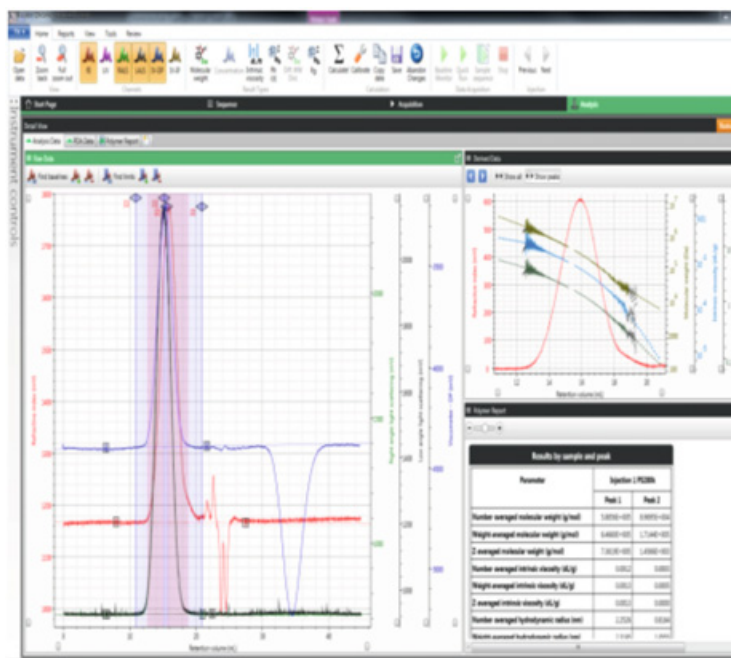
OMNISEC software v10, for OMNISEC REVEAL has been designed with you and your priorities in mind. The software is laid out to guide the user through setup, data acquisition and analysis in an intuitive workflow and its advanced user interface reduces training requirements for new users.

Key benefits of OMNISEC software v10 include:

- Intuitive look and feel
- Simple and customizable reporting to present only the data that is most important to you
- Overlay multiple injections and results quickly and easily
- Easy exporting of data
- 1-click from data to results.

VISCOTEK SEC-MALS 20

Absolute Molecular Weight, Molecular Size

The SEC-MALS 20 is a modular multi-angle light scattering detector that can easily be combined with any existing GPC/SEC system, for direct measurements of absolute molecular weight and size.
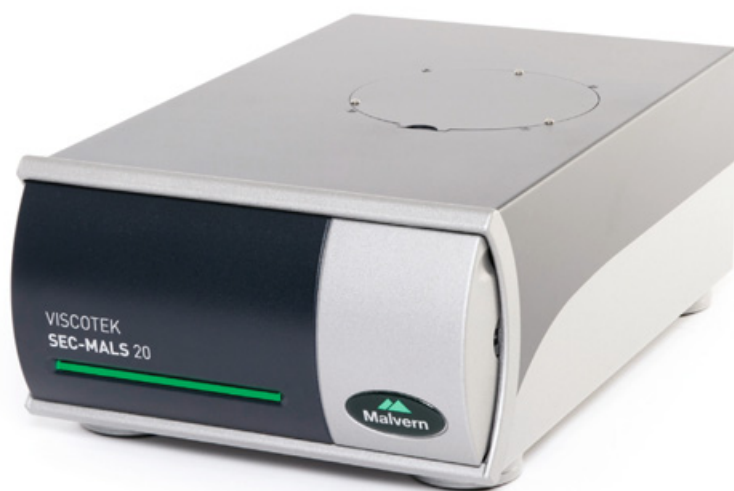
For proteins, the SEC-MALS 20 offers:

- Absolute molecular weight and oligomeric state

- Aggregate quantity, molecular weight and size (where applicable)

- Conjugation in samples such as PEGylated or membrane proteins (with two concentration detectors)

- 2nd virial coefficient A_2 (B_{22}).

For natural and synthetic polymers, the SEC-MALS 20 offers:

- Absolute molecular weight and molecular weight distribution

- Molecular size (Rg) for structural comparisons through conformation plots

- 2nd virial coefficient A_2 (B_{22}).
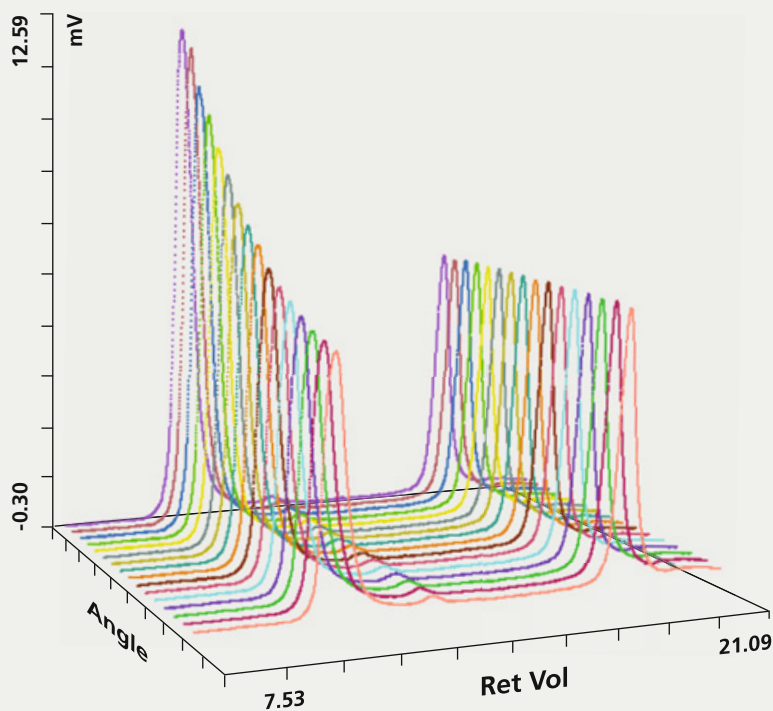
The key to the performance of the SEC-MALS 20 is the vertical flow cell with radial optics:

- Reduced detector noise at low angles

- Reduced need to clean the detector

- Fixed, constant and known measurement angles, regardless of solvent type

- One cell for all solvents means never having to switch or remove the cell

- Reduced band broadening and tailing compared with other MALS detectors, thanks to the low volume cell.

The overall result is minimized noise and the most accurate MALS data available.

MALS signals from an aggregated protein and monomer

ZETASIZER μV

Absolute Molecular Weight, Molecular Size

The dual capability Zetasizer μV offers unmatched versatility for protein analysis. It combines measurements of molecular weight by Static Light Scattering (SLS) with measurements of molecular size (Rh) by Dynamic Light Scattering (DLS). It can also perform both of these measurements in batch and GPC/SEC flow modes.

The detector is optimized for the measurement of smaller molecules that scatter less light, making it ideal for protein measurements.

SEC Mode

In flow mode, as a modular SEC-DLS detector, the Zetasizer μV measures the intensity of scattered light from which is calculated the absolute molecular weight of small macromolecules like proteins. It can also continually collect correlation functions to make measurements of Rh by DLS at the same time. As a GPC/SEC detector, the Zetasizer μV can measure:

- Absolute molecular weight and oligomeric state
- Aggregation levels
- Molecular size (Rh)
- Conjugate composition and molecular weight (with RI & PDA)
- 2nd virial coefficient A_2 (B_{22}).

Protein oligomers by RALS (green), RI (red) and Rh by DLS (black dots)
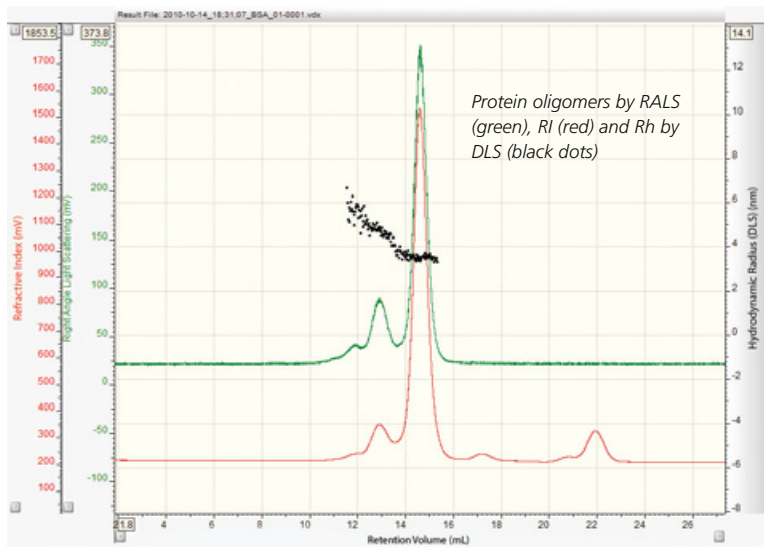
Chromatographic data from the Zetasizer μV in SEC mode

Batch Mode

In batch mode, using a cuvette, the Zetasizer μV can perform all of the DLS-based measurements you would expect from a Zetasizer instrument. Its temperature control allows you to perform the common temperature ramp, aggregation point, and stability experiments - all controlled using Malvern's Zetasizer software. In batch mode, the Zetasizer μV can measure:

- Hydrodynamic radius
- Aggregate detection
- Aggregation temperature studies.

DLS data from the Zetasizer μV in batch mode
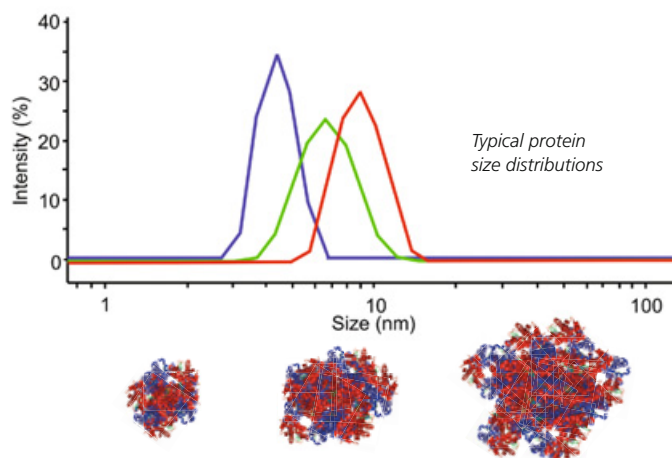
Typical protein size distributions

VISCOTEK **TDA**

Absolute Molecular Weight, Intrinsic Viscosity, Molecular Size, Concentration

The Viscotek TDA is a multi-detector platform for advanced GPC/SEC measurements.
It uses refractive index, light scattering and viscometer detectors to measure:

- Sample concentration
- Absolute molecular weight and molecular weight distribution
- Polydispersity
- Intrinsic viscosity
- Molecular size (Rh & Rg)

- Molecular structure/branching
- Mark-Houwink parameters
- Refractive Index increment, dn/dc
- 2nd virial coefficient, A_2 (B_{22})
- Copolymer and conjugate composition (with the PDA).

The integrated platform has these advantages:

- All columns and detector cells are housed in a single temperature-controlled compartment (up to 80 °C) for maximum baseline stability and solvent range
- Inter-detector tubing is minimized reducing band broadening and tailing.

The TDA as part of a complete system

VISCOTEK **TDA** TECHNOLOGY

Absolute Molecular Weight, Intrinsic Viscosity, Molecular Size, Concentration

The TDA's **refractive index (RI) detector** measures the concentration of almost any solute. The proprietary RI detector in the TDA has a robust flow cell keeping all detectors in series and maximizing their sensitivity.
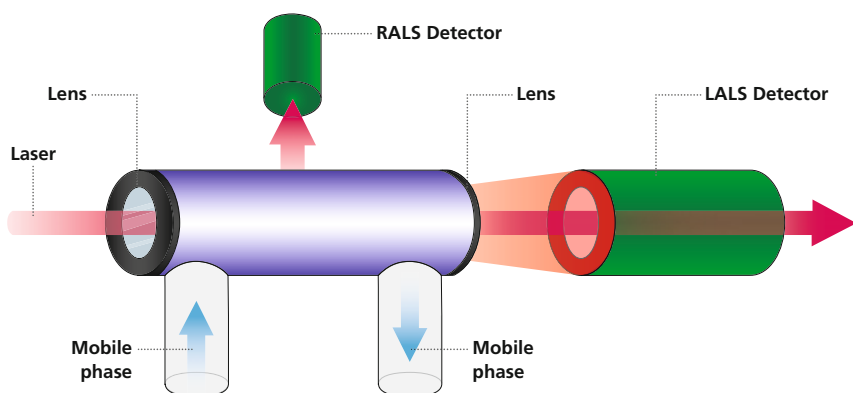
The **light scattering detector** inside the TDA combines the sensitivity of 90° Right Angle Light Scattering (RALS) with the accuracy of 7° Low-Angle Light Scattering (LALS). The software automatically chooses the best angle for the sample at every data slice. Its flow cell is just 18 µL minimizing band broadening.

Viscotek invented and patented the first **differential viscometer**. The TDA's digital transducers give a fast, sensitive response to viscosity changes, and 316 stainless steel construction means few sample limitations with salts or pH.

A RALS/LALS detector

VISCOTEK **RI** AND **UV** DETECTORS

Concentration

A modular differential refractive index detector for measuring sample concentration, which can be temperature controlled from ambient up to 55°C for improved baseline stability and is also compatible with sub-ambient measurements in a cold room or refrigerated cabinet. A 9 µl cell minimizes band-broadening.

The Viscotek UV detector is a variable, single-wavelength UV detector for the measurement of the concentration of UV active samples including proteins. It has a wavelength range from 190-500 nm. It has excellent sensitivity and a small footprint. Fiber-optic or stand-alone versions are available to allow integration into any system.
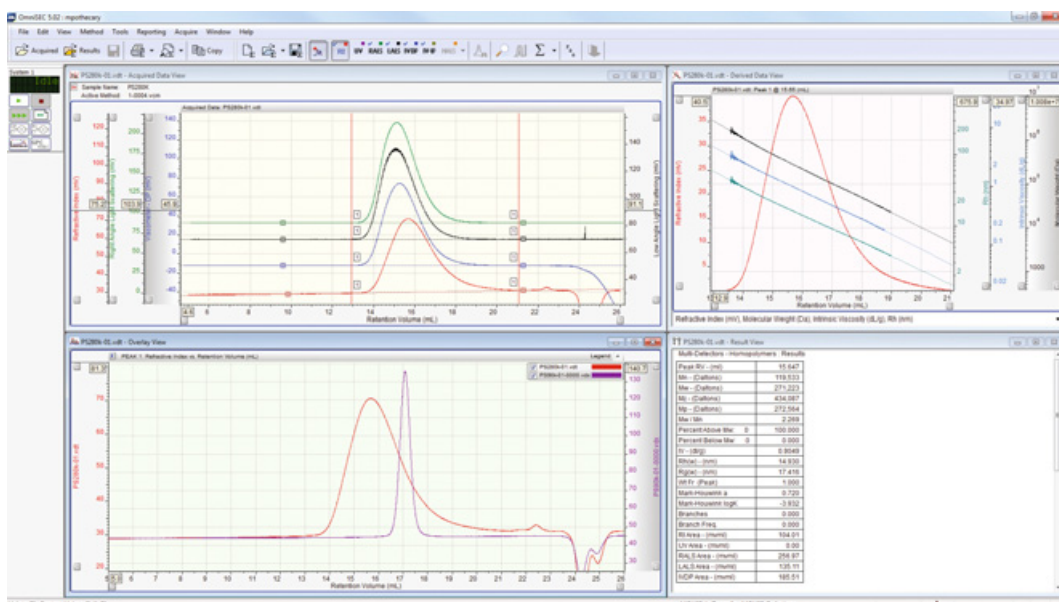
OMNISEC SOFTWARE v5

Control and analysis in a single package

OmniSEC software v5 is the GPC/SEC software package available for the Viscotek products.

With its clear user interface, OmniSEC software v5 is used to control Viscotek systems, acquire data from all Viscotek detectors and perform GPC/SEC data analysis.

With OmniSEC software v5 you can:

• Measure molecular weight and molecular weight distribution by light scattering or column calibration techniques
• Measure intrinsic viscosity and hydrodynamic radius. Mark-Houwink parameters and branching
• Be sure of the most accurate results thanks to proven band-broadening corrections
• Perform copolymer and conjugate calculations
• Overlay multiple chromatograms or results
• Reanalyze entire datasets in a few clicks using the Trend View
• Calculate dn/dc, dA/dc (extinction coefficient) and A_2 (B_{22})
• View all the angular data from a MALS measurement in a single plot with just one click of the mouse.
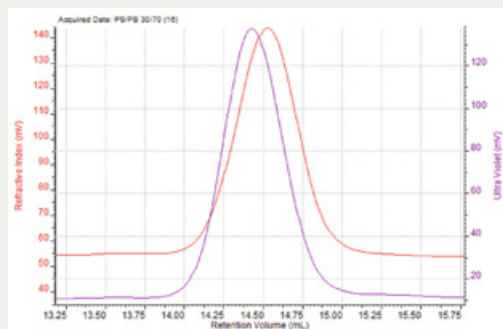
By combining RI and UV signals from a copolymer or conjugated sample, the software can calculate the concentration of each component and then measure the complex molecular weight and composition.

In this way, these samples can be fully characterized. Such samples include:

• Styrene/butadiene copolymers
• Conjugated protein samples such as PEGylated or membrane proteins.

Malvern Solutions: Advanced technology made simple - distributor details