

## 1.0 GPC Viscometry – collecting concentration and viscometry data

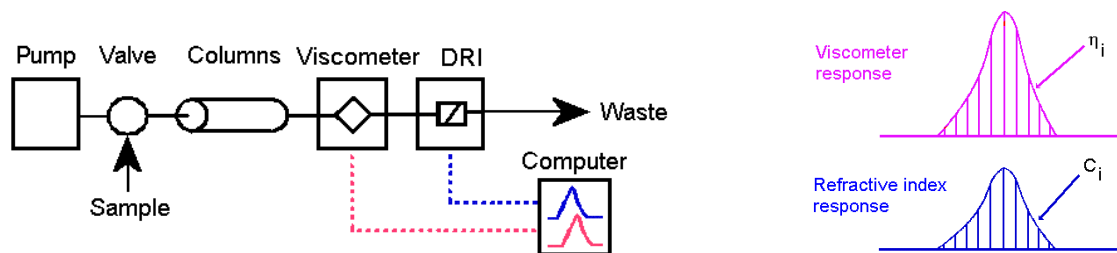
Using GPC Viscometry, **molecular weights** are determined using the universal calibration method. A plot of molecular size as  $\log(\text{molecular weight} \times \text{intrinsic viscosity})$  vs. **retention time** is constructed for a series of narrow standards based on the following relationships:

$$\text{Hydrodynamic volume} \propto \text{Molecular weight} \times \text{intrinsic viscosity}$$

Plot of  $\log(\text{MW} \times \text{intrinsic viscosity})$  vs. **retention time**  $\equiv$   $\log(\text{hydrodynamic volume})$  vs. **retention time**

Column separated and calibrated in terms of size therefore Universal Calibration obtained

### 1.1 Calculations by GPC Viscometry



Using the universal calibration calculations are performed by the following methods:

- The viscometry and concentration slices are obtained from the dual chromatogram of the sample
- For each slice of data on the concentration trace, the relative concentration is calculated based on the peak height and overall concentration of the sample
- For each slice on the viscometer plot, **intrinsic viscosity** ( $IV$  or  $[\eta]$ ) is determined from the specific viscosity and the concentration of the slice
- From the resulting retention time and **intrinsic viscosity** of the slice, **molecular weight** is calculated from the universal calibration plot
- Based on the calculated **molecular weight** values for each slice, the area of good quality data is fitted with a line and an extrapolation made to the high and low molecular weight regions of the data where the signal to noise is low
- The extrapolated **molecular weights** are used to calculate the molecular weight distribution
- From the **molecular weight** distribution and the **intrinsic viscosity** distribution, the **radius of gyration** ( $R_g$ ) distribution is calculated using the Flory-Fox relationship, assuming that the polymer has a random coil conformation

Using this method:

- **Intrinsic viscosities** are measured from the viscometer and concentration detector
- **Molecular weights** are calculated assuming that the sample obeys the universal calibration (pure size exclusion is obtained)
- **Radius of gyration** is calculated using a model for the polymer behaviour in solution

## 2.0 GPC Light Scattering – collecting concentration and light scattering data

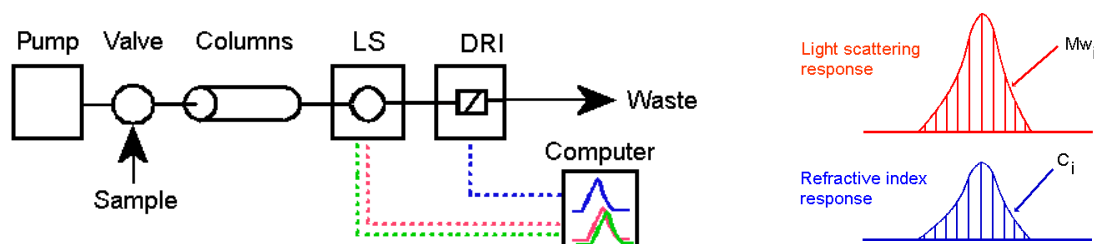
Using GPC Light Scattering, **molecular weights** are determined directly using the response of the light scattering detector with the following relationship:

$$R(\theta) = C M \left(\frac{dn}{dc}\right)^2 P(\theta) K(\theta)$$

Detector Response      Concentration X Mass      Specific Refractive Index Increment      Light Scattering Constant      Particle Scattering Function

Where the particle scattering function is a measurement of the change in the relative intensity of light scattering as a function of angle.

### 2.1 Calculations by GPC Light Scattering



Using GPC Light Scattering calculations are performed by the following methods:

- The light scattering and the concentration slices are obtained from the 15° and 90° light scattering and the concentration chromatogram of the sample
- For each slice of data on the concentration trace, the relative concentration is calculated based on the peak height and overall sample concentration
- For each slice on the light scattering chromatograms, **molecular weight** is determined from the response and the concentration of the slice
- Based on the calculated **molecular weight** values for each slice, the area of good quality data is fitted with a line and an extrapolation made to the high and low **molecular weight** regions of the data where the signal to noise is low
- The extrapolated **molecular weights** are used to calculate the **molecular weight distribution**
- From the ratio of the light scattering intensity at 15° and 90°, the **radius of gyration** is calculated from the particle scattering function
- From the **molecular weight** distribution and the **radius of gyration** distribution, the **intrinsic viscosity** (IV or  $[\eta]$ ) distribution is calculated using the Flory-Fox relationship, assuming that the polymer has a random coil conformation

Using this method:

- **Molecular weights** are calculated directly from the light scattering response, calculating the particle scattering function from the ratio of intensities at 15° and 90°
- **Radius of gyrations** are determined from the particle scattering function by comparison of the two angles but only if the molecule is over ca. 10 nm in size and the scattering intensity shows angular dependence

- **Intrinsic viscosity** is calculated using a model for the polymer behaviour in solution

### 3.0 GPC Triple Detection – collecting concentration, viscometry and light scattering data

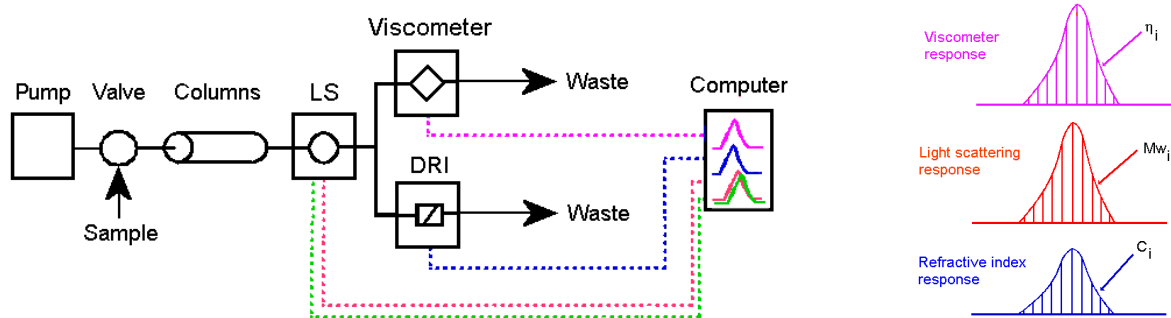
Using GPC Triple Detection, **molecular weights** are determined directly using the response of the light scattering detector with the following relationship:

$$R(\theta) = C M \left(\frac{dn}{dc}\right)^2 P(\theta) K(\theta)$$

Detector Response      Concentration X Mass      Specific Refractive Index Increment      Light Scattering Constant      Particle Scattering Function

Where the particle scattering function is a measurement of the change in the relative intensity of light scattering as a function of angle.

### 3.1 Calculations by GPC Triple Detection



Using GPC Triple Detection calculations are performed by the following methods:

- The light scattering and the concentration slices are obtained from the 15° and 90° light scattering and the concentration chromatogram of the sample
- For each slice of data on the concentration trace, the relative concentration is calculated based on the peak height and overall sample concentration
- For each slice on the light scattering chromatograms, **molecular weight** is determined from the response and the concentration of the slice
- For each slice on the viscometer plot, **intrinsic viscosity** ( $IV$  or  $[\eta]$ ) is determined from the specific viscosity and the concentration of the slice
- Based on the calculated **molecular weight** values for each slice, the area of good quality data is fitted with a line and an extrapolation made to the high and low **molecular weight** regions of the data where the signal to noise is low
- The extrapolated molecular weights are used to calculate the **molecular weight** distribution
- From the ratio of the light scattering intensity at 15° and 90°, the **radius of gyration** is calculated from the particle scattering function (the shape factor)

Using this method:

- **Molecular weights** are calculated directly from the light scattering response calculating the particle scattering function from the ratio of intensities at 15° and 90°
- **Radius of gyration** are determined from the particle scattering function by comparison of the two angles but only if the molecule is over ca. 10 nm in size and the scattering intensity shows angular dependence
- **Intrinsic viscosity** is calculated from the viscometer trace

#### **4.0 Comparisons between GPC Viscometry, GPC Light Scattering and GPC Triple Detection**

These three techniques can be used to determine 'absolute' **molecular weights** of samples, independent of the chemistry of standards used in the column calibration (GPC Viscometry) or independent of column calibration entirely (GPC Light Scattering and GPC Triple Detection).

The values of **molecular weight** can vary between the techniques for the following reasons:

- The viscometer and light scattering detectors respond to different properties of the polymer, the viscometer to molecular density the light scattering detector to size in solution, and so **molecular weights** calculated by these approaches will not necessarily have the same values
- **Molecular weights** calculated by GPC Triple detection may differ to those obtained by GPC Light Scattering as an iterative correction for the particle scattering function is carried out using the values of **intrinsic viscosity** from the viscometer and the Flory-Fox and Debye relationships – GPC Triple Detection is considered to give the most accurate molecular weight values

The values of intrinsic viscosity and radius of gyration can vary between the techniques for the following reasons:

- **Intrinsic viscosities** calculated by GPC Light Scattering without the use of a viscometer are obtained from the Flory-Fox relationship making assumptions about the conformation of the polymer in solution
- **Radius of gyration** values calculated by GPC Viscometry without the use of a light scattering detector are obtained from the Flory-Fox relationship making assumptions about the conformation of the polymer in solution
- Variations in **molecular weight** averages between GPC Viscometry, GPC Light Scattering and GPC Triple Detection will result in variations in the calculated **intrinsic viscosity** and **radius of gyration** distributions because the molecular weights are used in these calculations