# Use of High Speed/High Resolution Size Based Chromatographic Separation of Surfactants and Oligomeric Materials with Single Quadrupole Mass Spectrometry Detection

## Waters
### THE SCIENCE OF WHAT'S POSSIBLE.®

Author: Michael O'Leary, Jennifer Gough, Damian Morrison
Affiliation:  Waters Corporation, Milford, MA USA

## INTRODUCTION

Recent developments in polymerization processes have utilized a wide array of strategies.  The development has evolved from simple polymer chains to complex polymers capable of performing multiple functions within a single molecular chain.  As these new materials evolve their control and understanding has come under intense scrutiny utilizing a wide range of analytical technology ranging from chromatographic separation to advanced mass spectrometry.

Addressing the challenges of material characterization has often been focused on hyphenated detection techniques coupled with separation. This approach utilizes a concentration detector such as a refractive index (RI) detector as well as a viscosity detector and a multi angle light scattering detector and more recently the use of Mass Spectrometry (MS) detectors.

With the introduction of high speed-high resolution size based separation techniques, a novel approach to the design of the separation equipment including the separation column as well as the entire flow path are used to yield a high speed / resolution separation maintained from injection to detection with traditional detector options such as RI and UV detection.  However, the use of this high speed high resolution separation technique has seen limited pairing with MS detection due to the need to control material ionization and solvent matrix effects.

In this study the expansion of the APC approach to the size based separation is presented.  A single system control platform is evaluated to pair the chromatographic system to the MS detector system allowing for a high through put/ high resolution evaluation of size based separation of polymeric material while allowing for controlled MS ionization without interfering with the chromatographic separation.[1]

## METHODS

**Conditions for demonstration measurements:**

The test conditions are based on using a high speed, high resolution ACQUITY APC SEC system[1] coupled to a Waters SQD Detector 2 single quadrupole mass spectrometer.  The two detectors are managed by using a post column split flow at a fixed 1:1 ratio, (see figure 1).  Samples evaluated are PEG and Tergitol surfactants dissolved in the chromatographic system mobile phase.

**SEC Conditions**

- System                           Waters APC Core 1 System
- Eluent (PEG):              20/80 Methanol/Water  0.05% Formic
- Eluent (Tergitol):         95/5 Methanol/Water
- Flow rate:                     0.6 mL/min
- Injection volume:        10 µL
- Column Temp.:            40 ° C
- Columns                       2 Waters 4.6X150 mm BEH SEC 125 Å
- Conc. Detector            Waters ACQUITY RI detector @40 ° C
                                           (split between RI & Mass Spec)
- Split ratio                     Mass Spectrometer/RI  1:1

**Mass Detector Conditions**

MS system:                    Waters SQ Detector 2
Probe Type:                   ESI & IONSABRE II APCI
Ionization mode:          ESI⁺ ;

| | |
|---|---|
| Capillary voltage: | 3.0 kV |
| Desolvation temp.: | 500 °C ; |
| Source temp.: | 150 °C |
| Cone voltage: | 10 V; |
| Sampling rate: | 5 Hz |
| MS scan range: | 80 to 2000 *m/z* |

Ionization mode:          APCI⁺ ;

| | |
|---|---|
| Corona Current: | 1-4 uA (4 A for increased signal) |
| Desolvation temp.: | 500 °C ; |
| Source temp.: | 150 °C |
| Cone voltage: | 5-50V; (5V optimum for low charge) |
| Sampling rate: | 5 Hz |
| MS scan range: | 80 to 2000 *m/z* |

Waters Empower 3 FR2 Software for instrument control, data acquisition and chromatographic data processing.
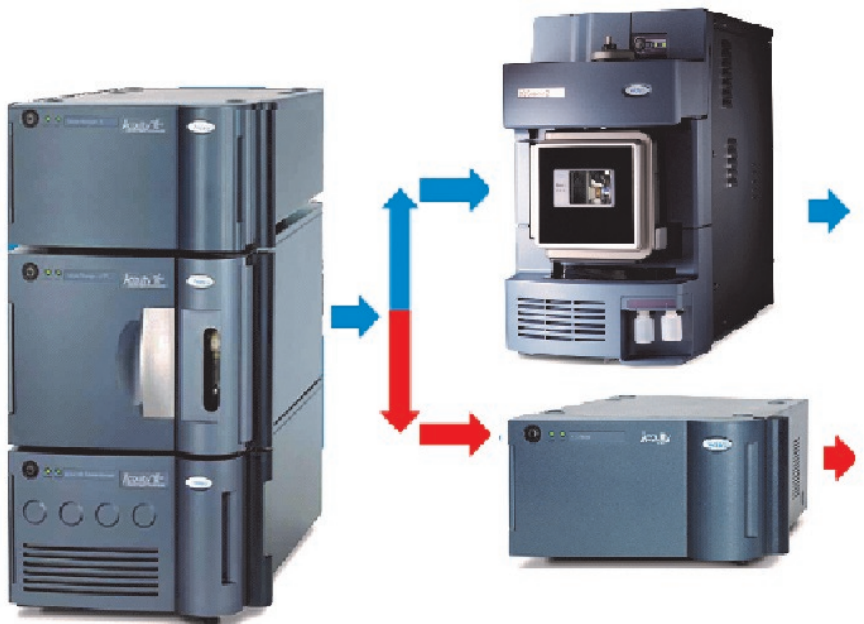
Figure 1.  Waters ACQUITY Core 1 APC system with post column split flow to ACQUITY RID and SQD 2 Mass Spectrometer
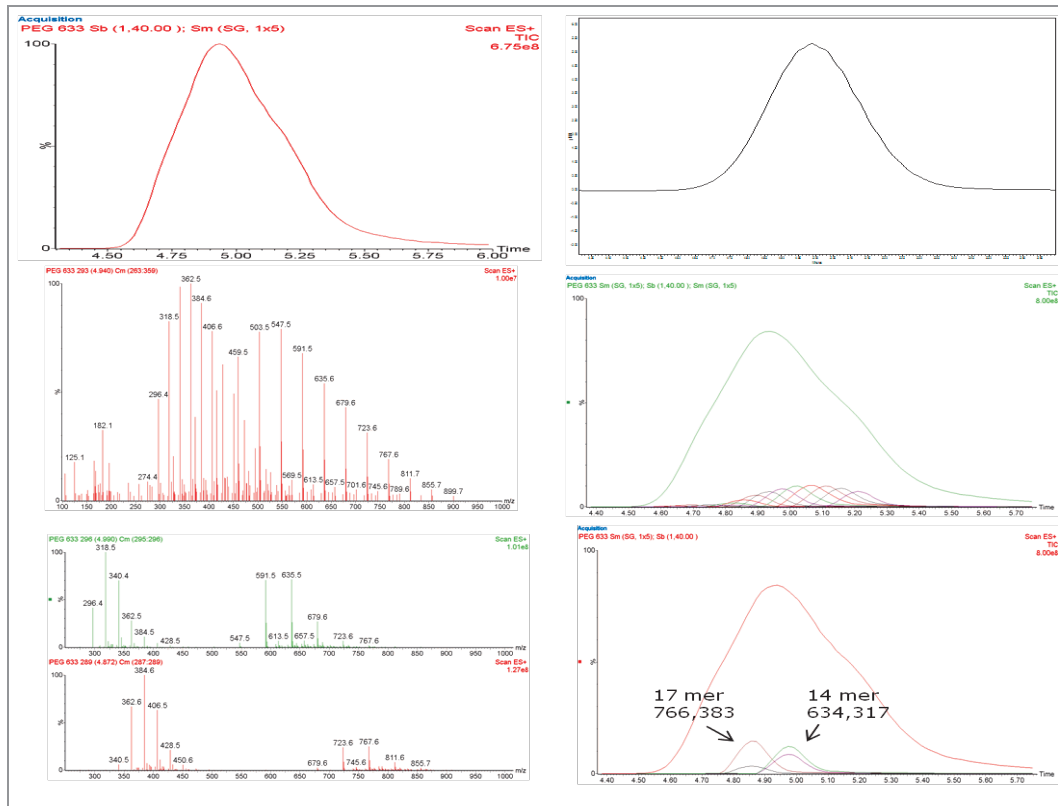
## RESULTS

Figure 2.  The top left image shows the Total Ion Chromatograph (TIC) using ESI conditions of a '633' Da PEG standard and the top right the corresponding RI signal from the same injection. The mid left spectra is the ESI collected across the entire size distribution showing the mass of each molecule separated by 44 Da (each EO unit or mer) as well as a second lower mass series separated by 22 due to the mass of each mer divided by the double charge (M/z).
The mid right chromatogram shows the TIC for the sample and the Extracted Ion Chromatogram (XIC) for the different chains lengths present in the 'narrow standard'.
The bottom left spectral set shows the spectra of two chromatographic (size) slices showing the impact of the charge 1 and charge 2 state of the sample.
The bottom right overlaid chromatogram set shows the distribution of the sample and that of the 17 and 14 mer chain length.  The overlapping peaks are due to the 2 charge states of the molecule.
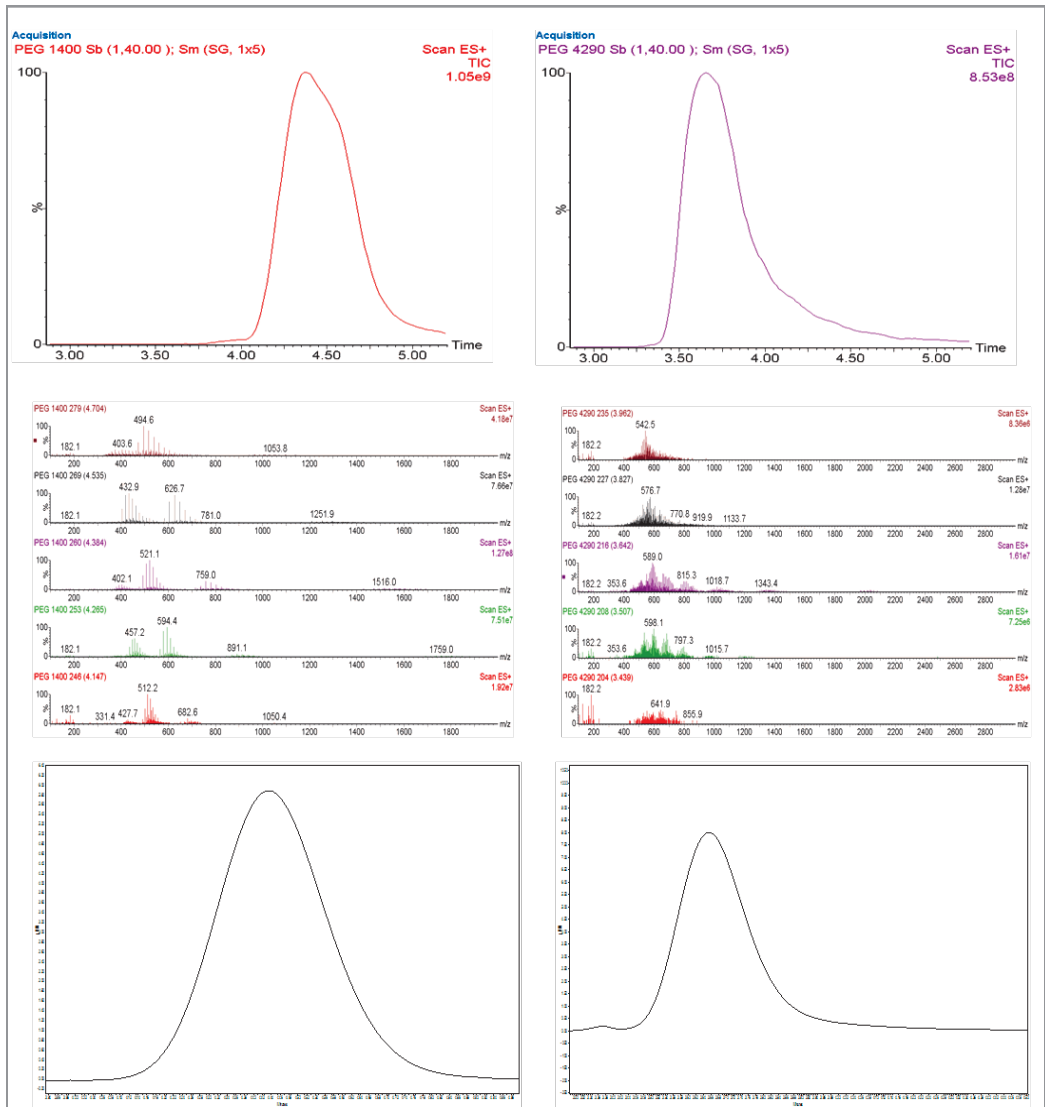
Figure 3. Top left image is the total ion chromatograms (TIC) using ESI conditions of a '1400' Da PEG standard, the mid left is the individual mass spectra as a function of retention time (size) showing +1, +2, +3 and +4 with increasing charging with chain length and the bottom left is the corresponding RI chromatographic trace for the polymer sample.
Top right image is the total ion chromatograms (TIC) using ESI conditions of a '4000' Da PEG standard, the mid right is the individual mass spectra as a function of retention time (size) showing +2, +3, +4, +5, +6 and +7 with increasing charging with chain length (note, +7 dominating) and the bottom right is the corresponding RI chromatographic trace for the polymer sample.
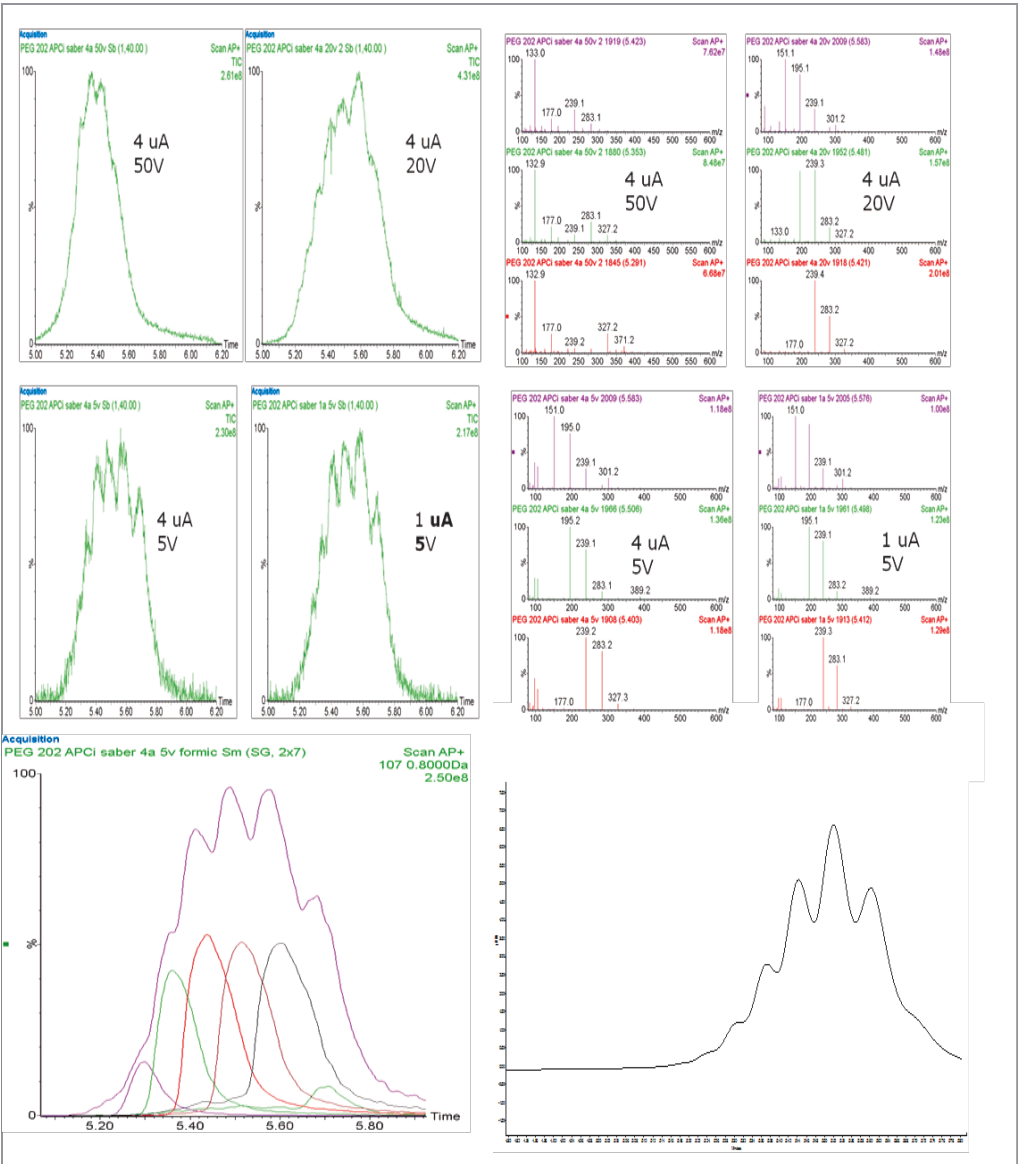
Figure 4. Top left set of images are the TIC of PEG '202' standard collected using Atmospheric Chemical Ionization (APCI) with varied corona current and cone voltage settings. Top right are spectra collected from each ionization condition extracted at the elution time corresponding to the signal maxima.
Bottom left image is the overlay of the smoothed TIC data of the PEG standard and the XIC spectra of the individual components in the mixture using the optimized cone voltage (5V) and corona current (4uA)settings.  The bottom right image is the corresponding RI chromatographic trace for the polymer sample.
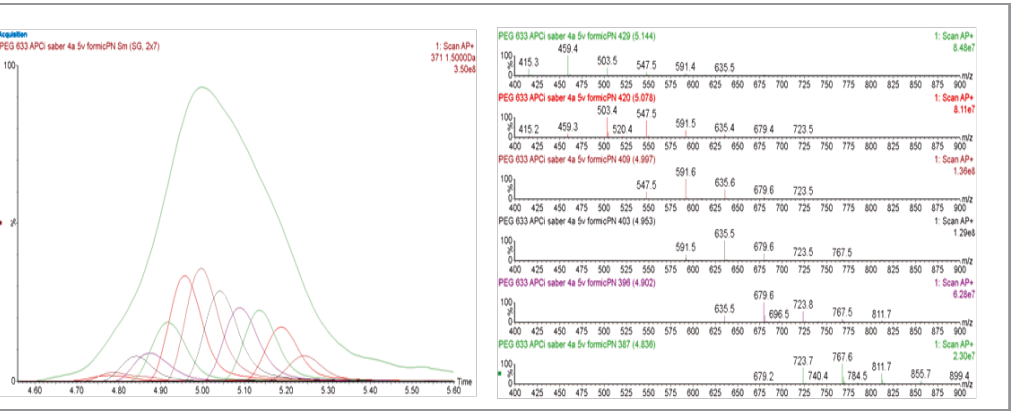
Figure 5. Left image TIC and XIC of '633' PEG using APCI. Right is the mass spectra of the components of PEG 633.
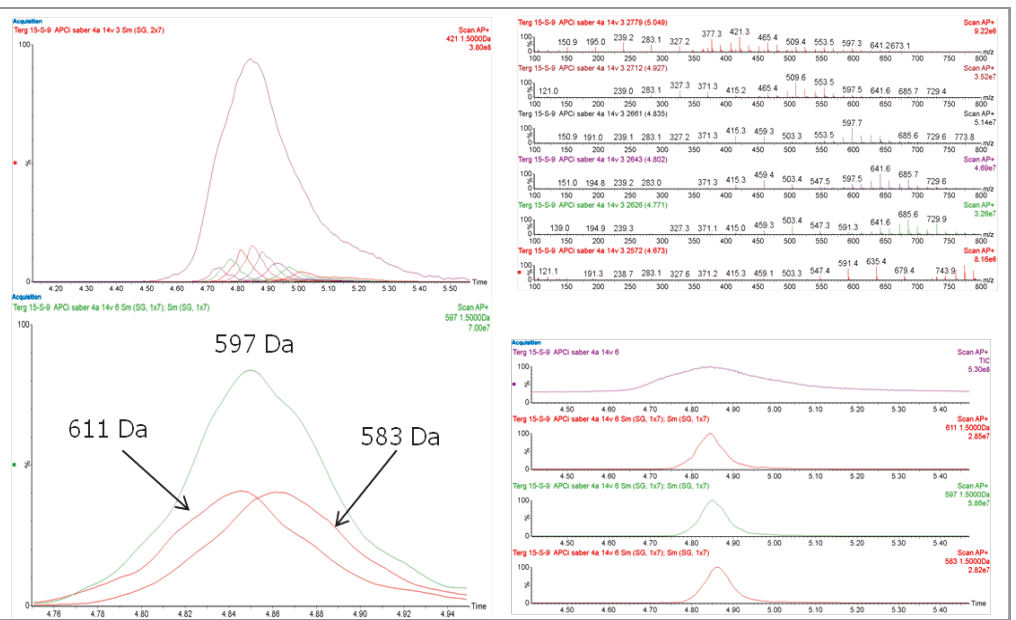
Figure 6. Top left image TIC and XIC of Tergitol[4] ($CH_3(CH_2)_{14}$ $(C_2H_4O)_5H$) using APCI. Top right are spectra collected from each ionization condition extracted at the elution time corresponding to the signal maxima.
Bottom left are the XIC of fixed EO content and varied CH2 length. Bottom right XIC relative to the full sample distribution

## CONCLUSION

- Using a single quadrupole  shows a potential for coupling MS to APC for rapid polymer analysis and MS optimization.
- Use of ESI yields rich data sets for interrogation but caution must be taken when evaluating the data due to charging effects with increasing chain length with increasing charge dominating the signal.
- Use of APCI allows for simplifying the mass spectral signal allowing for a simplified sample evaluation.

## DISCUSSION

In this study, two modes of ionization are evaluated as they relate to polymeric structure.  Using a high speed high resolution SEC separation each mode can be quickly optimized to assess the impact of each ionization mode.

In figure 2 we see the chromatographic profile of the PEG sample is available from the total ion chromatograph (TIC) using Electro Spray Ionization (ESI) showing good comparison with the more conventional RI signal for the polymer distribution.  Further we can examine the mass spectra of the sample and see the sample is made up of several different polymer chains.  Further investigation show the for each chain there can be at least two ions relating to the mass M and it charge z (M/z). showing both M/z for multiple charges can make interpretation difficult as shown for the 2 examples of the 17 and 14 –mer length chains.

In figure 3 two longer chain polymer samples are evaluated. Here the situation becomes more complicated as the degree of charging increases with increasing polymer length when using ESI.  Further as the polymer chain length increases the signal for the single charged chains all but disappears as the multiple charging species dominate.

In figure 4  we see the effects using Atmospheric Pressure Chemical Ionization (APCI).  Here we find the parent chain length will dominate the spectra at any point in the chromatogram as the charge or z will be 1.  The ionization mode is shown to be optimized to control any in source ionization by decreasing the cone voltage applied to the source resulting in a dominant signal for each molecule under investigation.

In figure 5 the APCI mode shows the same charge control for a longer chain PEG sample polymer with single charges dominating and the resulting M/z being equal to the mass of the polymer chain.

In figure 6 the APCI mode is used to evaluate a copolymer sample, Tergitol ($CH_3(CH_2)_{14}(C_2H_4O)_5H$).  Quickly the data set becomes more complicated due to the subtle structural changes in length of both the alkyl and ethylene oxide (EO) in the sample in some cases with nearly identical size.  The distribution of each molecule can be assessed using the mass of each chain in its single charge state showing unique composition.

### References

1. ACS Falll Meeting, Boston, MA, Polymer Division Paper ID 2253501 August 2015
2. Waters ACQUITY APC Product ordering guide, November 2014
3. Rapid Polymer Analysis with Atmospheric Solids Analysis Probe/ Mass Spectrometry (ASAP/MS), Waters Technology Brief 720003840en.
4. Tergitol ™ Manufactured by The Dow Chemical Co., Midland MI  Supplied by Spectrum Chemicals New Brunswick, NJ, USA

**TO DOWNLOAD A COPY OF THIS POSTER, VISIT WWW.WATERS.COM/POSTERS**