

Xevo G2 QTof System: High Resolution and Mass Measurement Accuracy for the Analysis of Complex Peptide Mixtures

GOAL

To utilize the Xevo™ G2 QTof for LC/MS^E analysis of complex tryptic peptide mixtures, illustrating the mass resolution and accuracy that can be obtained.

BACKGROUND

The identification of proteins in complex mixtures by nanoscale LC/MS is well accepted. More recently it has become apparent that high mass accuracy and resolution, particularly of the fragments produced by CID, provide significant specificity for identification purposes. By operating at high resolution and mass accuracy, more confident results can be obtained with a reduction in chimericity. In addition, false positive identifications are minimized due to the better mass accuracy and increased specificity. In this technology brief, we describe the analysis of complex peptide mixtures using the benchtop Xevo G2 QTof Mass Spectrometer, which operates at greater than 20,000 resolution (FWHM) with high sensitivity and mass accuracy.

The use of Xevo G2 QTof to analyze complex mixtures allows for protein identification and quantification with high resolution and mass accuracy.

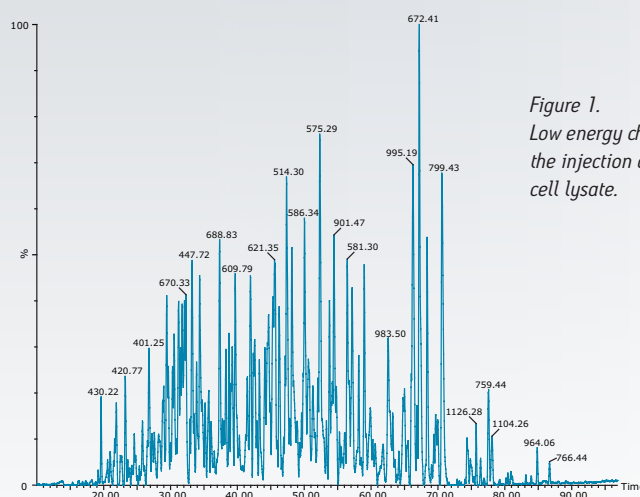


Figure 1.
Low energy chromatogram for the injection of 800 ng E.coli cell lysate.

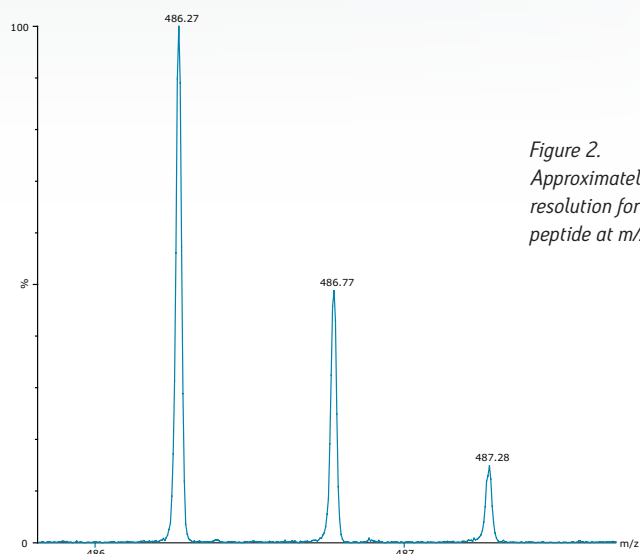


Figure 2.
Approximately 24 k (FWHM) resolution for a doubly-charged peptide at m/z 486.27.

THE SOLUTION

An 800 ng sample of digested *E. coli* cell lysate was prepared and analyzed using a nanoACQUITY UPLC® System coupled to a Xevo G2 QTof operating in resolution mode. The MS method used was LC/MS^E where the collision cell was switched between low and elevated energy during alternate scans acquiring peptide precursor information in the first function, and fragment ion information in the second function. A reference spray was sampled every thirty seconds to provide a lock mass correction. The low energy chromatogram is shown in Figure 1 and mass spectral resolution for an eluting peptide at m/z 486.27 is shown in Figure 2. Using ProteinLynx Global SERVER™ v. 2.4, data were processed and searched against a non-redundant *E. coli* database. To visualize the spread of mass accuracies of identified precursor and fragment ions, bar charts were created from search outputs, as shown in Figure 3 and Figure 4. The data shows that approximately 84% of identified precursors had mass errors within 2 ppm.

SUMMARY

The Xevo G2 QTof System provides high resolution and mass accuracy for the analysis of complex peptide mixtures, delivering a highly specific platform for protein identification and quantification.

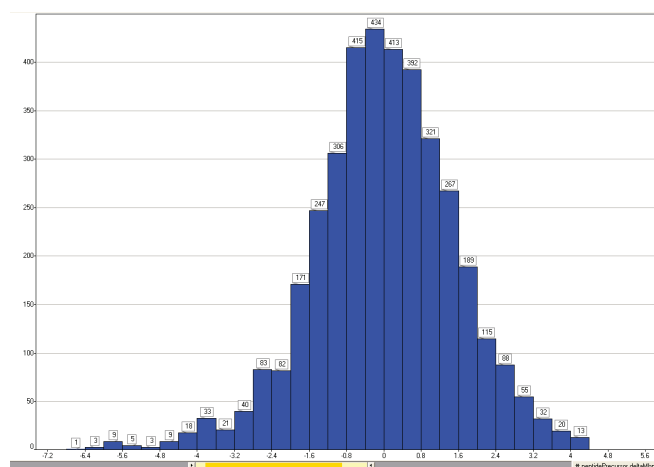


Figure 3. Mass accuracy for identified peptide precursors.

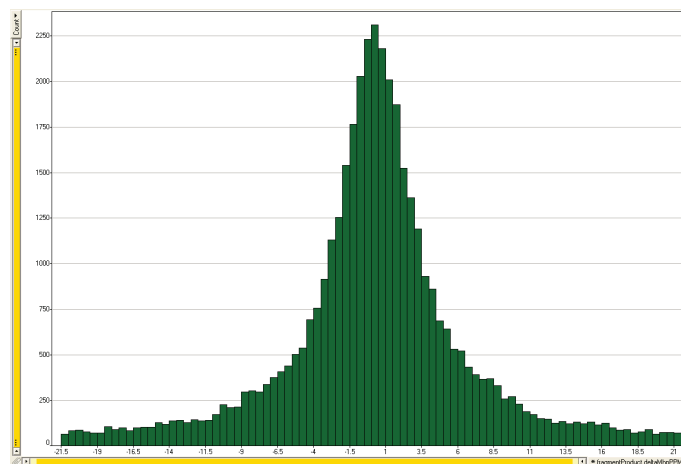


Figure 4. Mass accuracy for identified fragment ions.

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Waters Corporation
34 Maple Street
Milford, MA 01757 U.S.A.
T: 1 508 478 2000
F: 1 508 872 1990
www.waters.com

