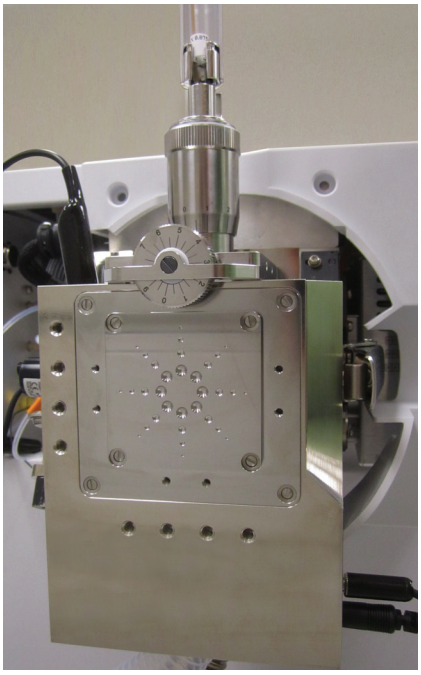
MAXIMIZE NANOFLOW LC/MS PERFORMANCE

Using the flexible, easy-to-configure Agilent nanoESI source



Agilent nanoESI source

Summary

The ability to detect and identify low abundance proteins is critical in advancing the field of proteomics and has led to the development of very small bore columns and nanoelectrospray (nanoES) interfaces to mass spectrometers. Agilent offers two sources designed specifically for maximizing nanoflow LC/MS performance: Agilent nanoESI source and the HPLC-Chip/MS interface.

While HPLC-Chip/MS technology integrates the nanocolumns and connections on a polymer chip to enable routine, robust and reproducible nanoflow LC/MS, the new Agilent nanoESI source is designed to provide the ultimate flexibility for experienced users. The new design easily accommodates columns of different lengths and inner diameter as well as multidimensional chromatographic configurations thus facilitating optimal performance for a wide variety of samples. The enclosed source both enhances safety and reduces background contamination from the laboratory. For Agilent accurate mass instruments, the nanoESI source incorporates a unique wick element for the continual introduction of reference mass ions enabling the highest mass accuracy, even under nanoflow LC/MS conditions.

Figure 1 shows that users can configure the emitter position for inline or orthogonal nanoES. All configurations accommodate the integrated camera and adjustable light source.

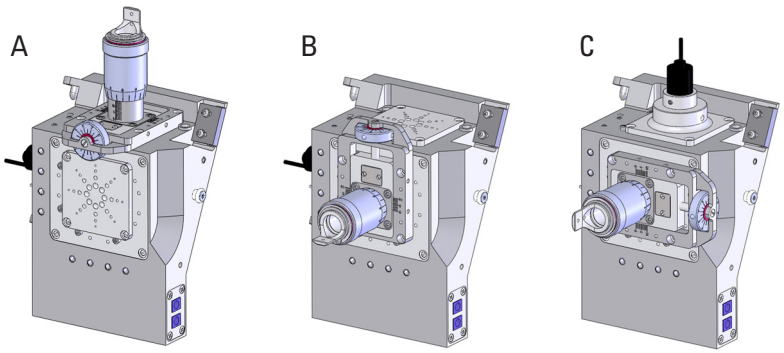


Figure 1. There are three possible configurations of the nanoESI source: orthogonal spray (A), inline spray with vertical adjustment (B), and inline spray with horizontal adjustment (C).

Online or offline nanoES

The Agilent nanoESI source can be used for offline or online nanoES, and provides easy connections for nanocolumns with integral (pulled tips) or separate emitters. Figure 2 shows the total ion chromatogram obtained for a 60-minute separation of a tryptic digest of an *E. coli* lysate performed using a 50 µm × 15 cm nanocolumn with an integral emitter. Protein identification was completed using Agilent Spectrum Mill proteomics software (version B.04) and results showed 6206 unique peptides and 913 proteins with a false discovery rate of 1.1 % (peptide) and 0.87 % (protein).

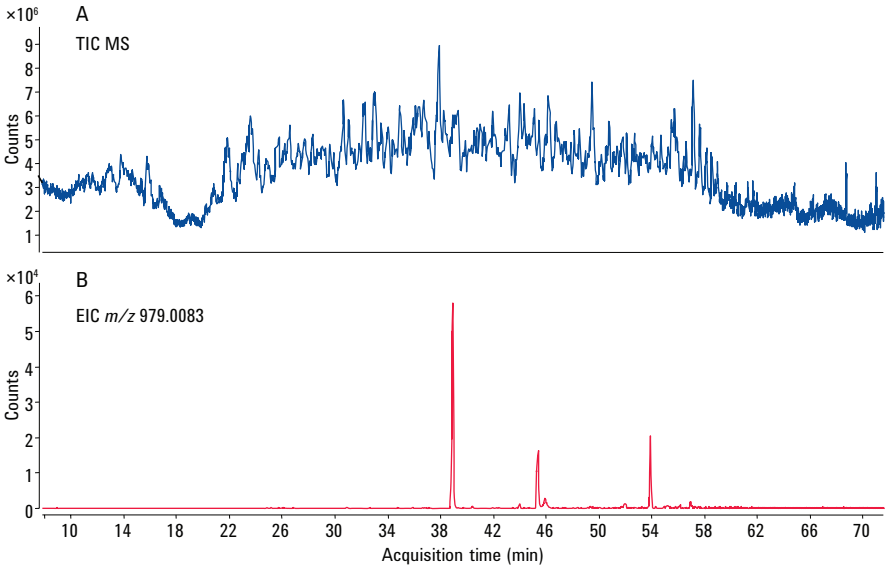


Figure 2. Total ion chromatogram for the separation of a tryptic digest of an *E. coli* lysate (A). A total of 500 ng was loaded on-column and a 60-minute gradient was used for the separation. Extracted ion chromatogram shows excellent chromatographic performance with good peak symmetry and a 10-second peak width at half-height (B).

nanoESI Source Highlights

- Compatible with all Agilent mass spectrometers
- Accommodates nanocolumns of any length or inner diameter
- Compatible with both online and offline nanoES
- Offers robust, precise positioning of the emitter
- Includes a camera and integrated light source for easy visualization of the emitter
- Supports either on-axis or orthogonal nanoES
- Accommodates reference mass addition on TOF-based mass spectrometers to obtain the best mass accuracy

Published in USA, September 7, 2012
5991-1041EN