# Bridging the micro-flow LC and nano-flow ESI: An emitter array ion source for sensitive and quantitative LC-MS analyses

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## Overview

- Chemically etched low dead-volume nano-ESI emitter array for effective coupling with micro-flow LC separation
- Multi-capillary MS inlet for efficient sampling of ions from emitter array ESI source
- Tandem ion funnel interface for high efficiency ion focusing and transmission

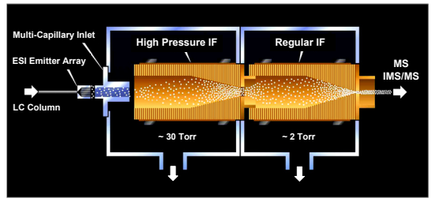
## Introduction

Operating electrospray in the low nL/min flow rate can significantly improve the analyte ionization efficiency, as well as reduce ion suppression and ionization bias in the electrospray ionization (ESI) process. However, most liquid chromatography-mass spectrometry (LC-MS) based analyses operate at much higher flow rate than required for nano-electrospray operation. This flow rate mismatching generally limits the utility of nano-ESI in most LC-MS applications. We present an ESI source and MS interface that effectively combine micro-flow LC with nano-ESI to achieve high sensitivity in LC-MS analysis. The key elements in the new ESI source include an ESI emitter array[1] with optimum geometric configuration,[2] matched MS sampling inlet array,[1-2] and a tandem ion funnel interface.[3]

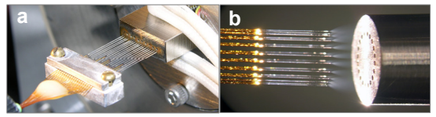
The integration of micro-flow LC and nano-ESI was achieved by splitting the LC effluent (~μL/min) among an array of nano-ESI emitters.[4] To effectively sample the high ion current generated by the emitter array, a heated multi-capillary was used as the MS inlet. This design allows each ESI emitter to be aligned with an individual heated inlet capillary. The increased gas throughput by the use of a multi-capillary inlet was effectively handled by a tandem ion funnel interface that also allowed the large ion current exiting the multi-capillary inlet to be transmitted into MS with high efficiency.

## Methods

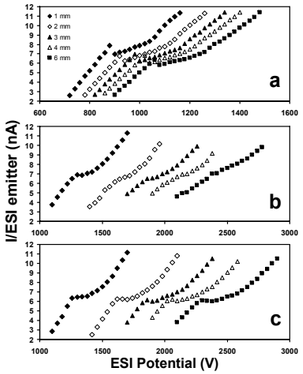
### ESI emitter array ion source and multi-capillary inlet/tandem ion funnel interface for MS




### Chemically etched (a) linear emitter array, (b) radial emitter array with better inter-emitter electric field uniformity [4] and their corresponding matched multi-capillary inlets

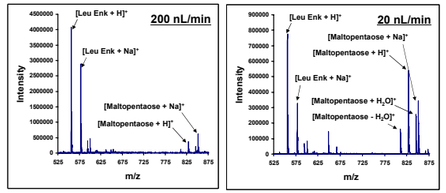



Electrospray current vs. voltage at various emitter–counter electrode distances for (a) a single emitter, (b) a linear array of 19 emitters, and (c) a circular array of 19 emitters. The flow rate was 50 nL/min in (a) and 950 nL/min in (b) and (c). The solution was 1:1 $H_2O$:MeOH + 0.1% HOAc.



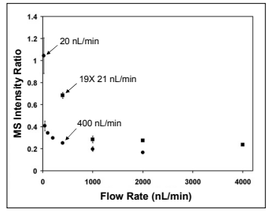

## Results

### Reduced charge competition/ionization bias in ESI process using ESI emitter array ion source

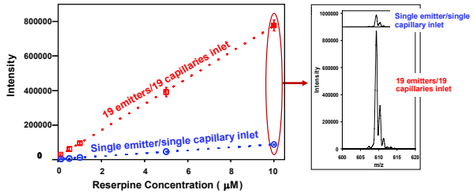



Sample: Equal molar (10 μM) leucine enkephalin and maltopentaose in 1:1 MeOH/10 mM aqueous ammonium acetate; ESI: 1.2 kV, ~1.5 mm gap; MS: LCQ ion trap MS

MS intensity ratio of maltopentaose and leucine enkephalin intensities vs. flow rate for a single emitter and an array of 19 emitters



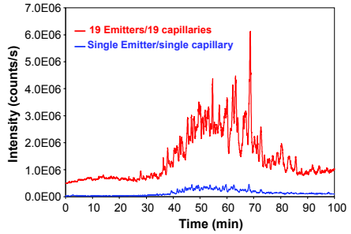

### Sensitivity improvement using emitter array ion source and multi-capillary inlet



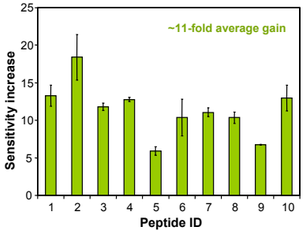

MS sensitivity using 19 ESI emitters/19 (430-μm) capillary inlet interface was compared to a standard single ESI emitter/single capillary inlet interface on MSD 1100 single quad MS. Total infusion low rate 1 μL/min. Solvent: 1:1 MeOH:$H_2O$ + 1% acetic acid.

### LC-MS analyses of tryptic digest of protein standards spiked into depleted human plasma (1 mg total sample)[2]

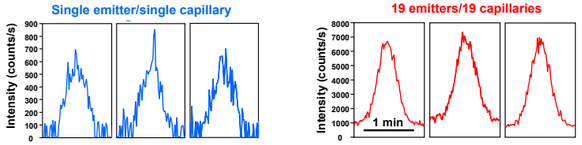
19-emitter/19-capillary interface and a single emitter/single inlet interface
LC: 65 cm long, 150 μm i.d. fused silica column packed with 5 μm porous C-18 particles
Total flow rate: ~1.8 μL/min




Total ion chromatograms of the LC-MS analyses

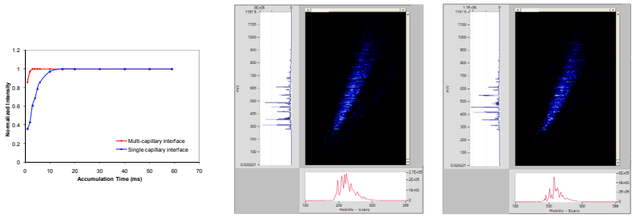



Relative sensitivity improvement for ten randomly selected peptides resulting from multi-emitter/multi-inlet interface




Extracted ion chromatograms (EICs) of LPSEFDLSAFLR, a low-abundance tryptic peptide from *E. coli* β-galactosidase spiked into human plasma, from 3 replicate LC-MS analyses using 19-emitter/19 capillaries and single emitter/single capillary configurations for both peak width and sensitivity comparison.

### Significantly reduced ion accumulation time using multi-capillary/tandem ion funnel interface on an IMS/TOF MS




a) Normalized MS signal intensities at different ion funnel trap accumulation times for multi-capillary and single capillary interfaces; b) IMS/MS data using single capillary interface at 50 ms accumulation time; c) IMS/MS data using multi-capillary interface at 3 ms accumulation time sensitivity. Total infusion flow rate 100 nL/min. Sample: BSA digest at 0.1 mg/mL in 1:1 MeOH:$H_2O$ + 1% acetic acid.

## Conclusions

- Micro-flow LC can be effectively split into nano-flow ESI using emitter array for high ion current and reduced charge competition/ionization bias in ESI process
- Radial ESI emitter array improves inter-emitter electric field uniformity
- Multi-capillary inlet improves significantly the ion sampling efficiency through ESI-MS interface
- Tandem ion funnel interface efficiently focuses and transmits ion current in the ESI-MS interface region
- Order of magnitude sensitivity improvement has been observed using the emitter array/multi-capillary tandem ion funnel interface on different MS platforms

## Acknowledgements

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