Cone-jet mode electrospray Ionization in micro- and nanoflow regimes by emitter surface manipulations

Sau Lan Staats, Anna Stoltzfus, Eliana McCray and Andris Suna, Phoenix S&T, Inc. , Chadds Ford, PA 19317 USA

Overview

The cone-jet mode spray in nanospray-MS produces enhanced sensitivity which is >10x of conventional ESI. NanoLC-MS requires fused silica columns and emitters which are fragile and spray stability and clogging disrupt analyses regularly. Two new spray emitters, one metallic and one plastic, have been under development for providing cone-jet mode spray for nanoLC-MS and microLC-MS. We have shown that the metal emitter could produce nanospray MS sensitivity with microspray (2-10 uL/min) flow-rates, opening up the possibility that robust nanospray-level sensitivity may be accessible to fields outside of proteomics: lipidomics, metabolomics and ultimately clinical testing. The report presents studies on interfacial interactions (contact angle properties) of different materials used as spray emitters to further probe the spray mechanism.

Introduction

The cone-jet mode spray in nanospray-MS produces enhanced sensitivity which is >10x of conventional ESI. NanoLC-MS can be intimidating because the fused silica columns and emitters are fragile and the spray instability and clogging disrupt analyses regularly. Recent experimental evidence[1] shows that microspray (2-10 uL/min) could produce nanospray-level sensitivity using metal emitters and columns that are very robust, potentially making nanospray-level sensitivity in LC-MS accessible to researchers in fields including lipidomics and metabolomics. We present a novel approach to create a cone-jet mode emitter suitable for flow-rates of 100 uL/min and beyond by optimizing the interfacial interactions (contact angle properties as opposed to the frequently studied surface tension of the sprayed buffer) of different materials used as spray emitters.

The theoretical framework with which we started the investigation was the well-known semi-empirical equation[2] for the spray voltage on-set:

$$V_{on} \sim \sqrt{\frac{r_c \gamma \cos\theta}{2\varepsilon_o}} \ln\left(\frac{4d}{r_c}\right) \qquad \text{Eq. (1)}$$

where r_c is often treated as the outside radius of the capillary used as the spray nozzle, ε_o is the permittivity of vacuum, γ is the surface tension of the solution being sprayed, d is the distance between the counter-electrode and the tip of the capillary, and θ is the half angle of the Taylor cone angle (49.3˚). This equation has been adequate to describe qualitatively experimental observations.

We have shown in previous studies that the interfacial interaction term may constitute a part of the proportionality constant of the above equation. The wettability of the emitter surface by the buffer being sprayed, especially when the buffer is mostly aqueous is a likely parameter for affecting the voltage threshold. The theoretical modeling and the experimental spray observations were designed to further test the validity of this assumption by introducing new surface modifications to the metal emitters.

Method

A. Emitter Design

Spray emitters made of four different materials were studied. The following table gives the contact angles with water from our own measurements

Emitter materials	Contact Angle (degrees)
Fused silica	~30
Polished stainless steel	~80
Gold-coated metal	~82
E-beam deposited Al_2O_3 on polished SS	~70
Plastics –PEEK, Polypropylene	>>90 (hydrophobic)

The actual contact angles depend on many factors: the roughness and cleanliness of the surface, the purity of the water, etc. We believe that our highly-polished metal surfaces created slightly more hydrophobic contact angles than those reported in the literature[3]. We focused on the trend instead of the actual values of the contact angles.
The emitter openings were 25, 50 um, 60 um and 100 um for the metal emitters, and 20 um for the fused silica and PEEK emitters.
If the metal emitters were tapered, the o.d of the spray emitter openings were ~100 to ~120 um tapered from a capillary o.d. of 300-360 um.

The e-beam deposited gold coatings were from ~20 to about 1000 angstroms thick and that of the Al_2O_3 was also 1000 angstroms thick. The e-beam deposition was carried out at UDNF at the U. Delaware.

The PEEK tubing had a 20 um i.d. and 360 um o.d. The emitter opening was slightly tapered. The polypropylene emitter is in the form of a conical micro- injection-molded nozzle with a ~30 um i.d. and 100 um o.d..

B. Spray conditions

Flow rates from submicroliter/min to > 200 uL/min were obtained from a syringe pump or the split flow of an LC (Shimadzu LC-6A) . The spray buffers used were

99.8% water/0.2% Formic acid
50/50 Methanol/Water
100% Methanol

A CCD camera with a zoom lens captured the spray images which were digitized and stored.

Results

A. Modeling

1. Spray voltage threshold vs. contact angle

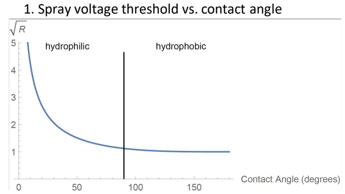


Figure 1: A plot of the square root of radius of curvature of the liquid tip against contact angles. It shows that the somewhat less hydrophilic solvent can result in a significant lowering of the critical voltage for electrospray. It is reasonable to assume that this voltage is proportional to the square root of the radius of curvature *R* the liquid has before any voltage is turned on. There is also a log[d/R] factor where d is the distance to ground, but for very large d this is insensitive to R. R was calculated for a drop of fixed volume, as a function of the contact angle when this drop sits on a plane surface.

2. Shape of the static liquid surface at the emitter opening as a response to surface interactions

The shape of the static liquid surface of liquid exuded from a spray emitter prior to the application of an electric field is shown below. Fig 2 a and b apply to any liquid interface and is based on the Young equation[3], which equates pressure to curvature. This shape which is determined by the nozzle geometry, contact angle, and the volume of liquid outside the emitter opening can predict, at least qualitatively, some properties of electrospray. The onset of electrospray can be understood to occur when the curvature of this surface, when modified by an electric field, exceeds a critical value. When the field is applied, charge on the surface gets concentrated more the larger the curvature which then in turn increases curvature further. Roughly, the threshold field for spray onset will be proportional to the difference between the critical curvature and the zero-field curvature. As a function of the exuded volume, spray will most likely start when the zero-field curvature is maximum.

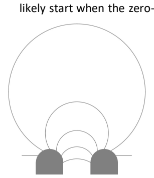


Figure 2 (a)

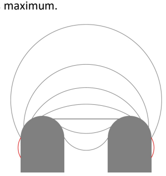


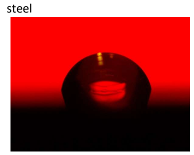
Figure 2 (b)

Figure 2 (a) illustrates the hydrophobic case, for a contact angle of 120 degrees. The Features to note: the maximum curvature occurs when the liquid contacts the inside surface of the emitter opening. In contrast, in Figure 2 (b) which illustrates the hydrophilic case of a contact angle of 30°, the maximum curvature occurs at the **outer** surface of the nozzle opening. A critical maximum volume is reached, beyond which point a high curvature overflow ring is formed (red) .

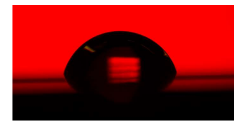
B. Contact angle measurements

The contact measurements were made with the contact angle measurement apparatus in the Advanced Materials Characterization Labs (AMCL) at the University of Delaware.

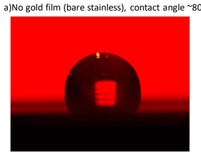
Fig.3.: Images of the water drops on e-beam deposited gold films on polished stainless steel



a)No gold film (bare stainless), contact angle ~80°



b) 1000 Å Alumina film, contact angle: 708°



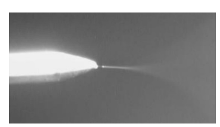
c) 1000 A gold, contact angle ~82°

d) Glass slide, contact angle ~30°

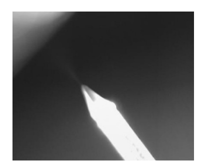
The contact angle measurements of e-beam-deposited thin gold films on highly polished stainless steel plates show that the gold film tends toward hydrophobicity when compared to the native stainless steel surface, which is a surprise at first glance as there is a notable amount of evidence[4] that water wets (~0° contact angle) a gold surface. However it has been shown[5] that the contact angle of water on gold films with patterned microscale structures may turn hydrophobic (>90°) as we also observed in a clear-room sputtered gold surface which showed rough morphology through SEM. The drop image on a glass slide shown in Fig. 3d) showed that the contact angle measurement apparatus was working properly. The ~80° (as opposed to the ~72° as reported in the literature) of the bare stainless steel surface may have been due to the fact that the stainless steel plate had a perfect mirror finish. The contact angle of a drop of water on a highly polished Ni surface was measured to be ~87° with this apparatus, agreeing with the difference of the contact angles between these two surfaces in the literature. The alumina surface was slightly more hydrophilic than the solid metal surfaces.

C. Electrospray Observations

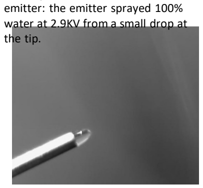
Figure 4: Water drop at emitter opening: Hydrophilic vs hydrophobic emitter surfaces



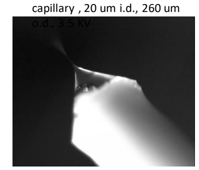
a. Al_2O_3 film on a tapered metal emitter: the emitter sprayed 100% water at 2.9KV from a small drop at the tip.



b. Flat-cut fused silica capillary , 20 um i.d., 260 um o.d., 3.5 KV



c. Flat-ended stainless steel emitter, 60 um i.d., 280 um o.d. 3.5 KV



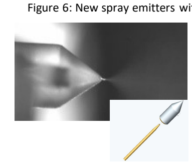
d,. Partially tapered PEEK tubing 20 um i.d., 360 um o.d , 4 KV.

Figures 4 a -d show qualitatively that the water droplet shapes were consistent with the calculated models in Figures 2a and 2b. Th alumina surface, being slightly more hydrophilic than polished metal surfaces, was capable of cone-jet mode spray but the spray was from a small drop. In Figure 4b where the emitter surface was very hydrophilic, the droplet coming out of the emitter opening wetted the surface surrounding the opening immediately and it became impossible to have a good cone-jet spray directly at the opening. It is interesting to note that the droplet behavior in the stainless steel emitter in Figure 3c resembled that of the PEEK emitter where the water droplets did not flow over the edge of the emitter opening even though a metal surface is generally considered hydrophilic. For Figures 4 c, a finite voltage insufficient to induce spray was applied to each droplet, whereas in Figures 4 a, b &d, the cone-jet mode spray was observed at the applied voltage.

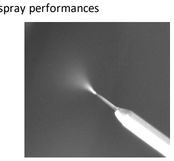
Fig. 5 : Effect of surface film modification

We noticed that the surface film on the metal emitter could change with use. For example, the gold-coating on the emitter could over time become more hydrophilic and lose its cone-jet mode spray ability. A "regeneration" of the film would become necessary. Sonicating the emitter in dilute HNO3 usually succeeds in restoring the cone-jet mode spray for a period of time. However, applying the same procedure to an alumina film would render the film porous as shown in the figure here. a drop of methanol emerging from the tip "wrapped" around the tip because the film had been made porous by the acid.

Figure 6: New spray emitters with previously inaccessible spray performances



a) A polypropylene micro-injection –molded poly propylene nozzle spraying methanol at 3 KV at 800 nL/min. This nozzle is non-clogging and can be connected to a 360 um o.d. capillary without fitting



b): The tapered metal emitter with reduced wetting sprayed methanol in cone-jet mode at 160 uL/min and 3.5 KV

Summary and Conclusions

- We have discovered that the interfacial interactions between the buffer and the emitter surface may be manipulated to facilitate the cone-jet mode spray by reducing the voltage threshold for the Taylor cone instability.
- A major contributor to the interfacial interactions may be characterized by the contact angle between the buffer and the emitter surface: the more hydrophobic is the emitter material, the more likely is the formation of the cone-jet mode spray.
- Metals such as stainless steel can be induced to exhibit the cone-jet mode spray of an aqueous buffer by rendering their emitter surfaces more hydrophobic either mechanically or electrochemically.
- For metals, surface roughness increases wetting. Therefore a metal with smoothed surfaces will likely produce better electrospray than a rougher surface.
- By tapering the emitter in Figure 6b above, a cone jet mode spray of methanol at 160 uL/min. Flow rates as high as 260 uL/min have been observed to produce cone-jet mode sprays.
- This discovery opens up the possibility of enhanced sensitivity at conventional ESI flow rates of >100 uL/min. We have already demonstrated[1] that this emitter with reduced wetting achieved nanospray sensitivity at 2-5 uL/min in both positive and negative ion spray. Some aspects of this emitter is patented while others are patent pending.
- A thin film of gold coating on a metal emitter can potentially lengthen the life of the hydrophobic metal emitter.
- A polypropylene micro-injection-molded spray nozzle specifically designed for nanoLC-MS is near completion. The nozzle in a chip-array format had been shown[6] to be non-clogging and sprayed superbly during the entire gradient.
- We are testing these emitters for mass spectrometry performance under different experimental conditions in collaboration with Papa Nii Asare-Okai, PhD at the Mass Spectrometry Facilities at the University of Delaware.

References:

1. H-K Lim, proceedings of the ASMS conference, 2017
2. Kebarle, P; Ho, Y., Cole, R. C., Ed. John Wiley & Sons, Inc. New York, **1997**, 3-63.
3. K.W. Bewig, W. A. Zisman, J. Phys. Chem. *69,* 4238-4242, 1965
4. R. J. Good, J. Adhesion Sci. Technol., *6*, 1269-1302, 2012
5. M. E. Abdelsalam et al, Langmuir, *21*, *1753-1757, 2005*
6. M Myers, SLT Staats, I. Ajagba, Proceedings of the ASMS Conference, *2006*

Acknowledgments: The work reported here was partially supported by an NIH (NIGMS) SBIR grant # 1 R43 GM133343-01
We wish to thank PapaNii Asare-Okai, PhD and Kojo Otchere-Addo of the Mass Spectrometry Core Facilities of the University of Delaware, Scott McCracken and Iulian Codreanu of the Nanofabrication Facilities for their help with the e-beam deposition, and Gerald Poirier of the Advanced Materials Characterization Labs, both at the University of Delaware.