

Novel Characteristics of Probe Electrospray Ionization

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Probe electrospray ionization (PESI) is a recently developed ionization technique which uses a solid needle or wire as sampling probe and ESI emitter instead of capillaries. The needle or wire is moved up and down along a vertical axis using a custom made linear actuator system. When the needle is at the bottom position, the tip of the needle is adjusted to touch the surface of the liquid sample. When the needle or wire is moved up to the highest position, a high voltage of about 2.0-3.0 kV was applied to it. The distance between the top and bottom position of the needle is about 10 mm. The ions generated from the electrospray are sampled through the ion-sampling orifice with a diameter of 0.4 mm into the vacuum chamber and mass analyzed by an orthogonal time-of-flight mass spectrometer. Figure 1 shows the schematic diagram of probe electrospray ionization (PESI/MS) system.

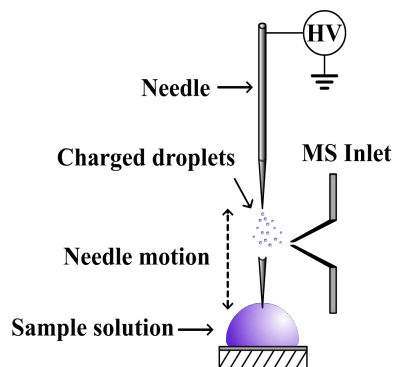


Figure 1: Schematic diagram of probe electrospray ionization mass spectrometry (PESI/MS).

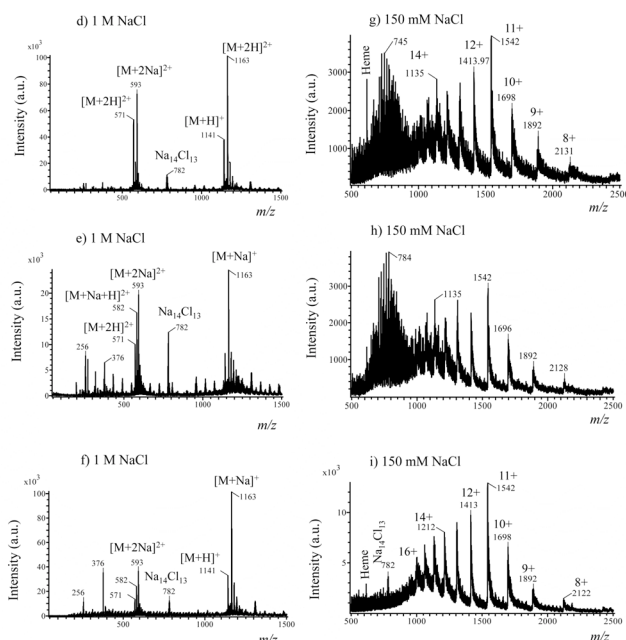
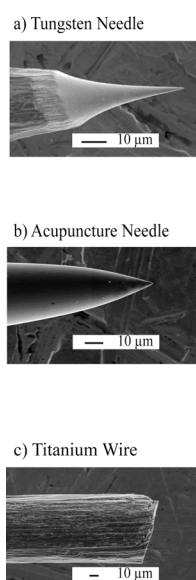


Figure 2. SEM images of **a)** tungsten needle, **b)** acupuncture needle (stainless steel) and **c)** titanium wire. **d)-f):** Positive ion PESI mass spectra of 10^{-5} M gramicidin S in the solution that contains 25% (v/v) MeOH, 1% (v/v) HAc and 1 M NaCl, obtained by tungsten needle (d), acupuncture needle (e) and titanium wire (f). **g)-i):** Positive ion PESI mass spectra of 10^{-5} M myoglobin in the solution that contain 25% (v/v) MeOH, 1% (v/v) HAc and 150 mM NaCl, obtained by tungsten needle (g), acupuncture needle (h) and titanium wire (i).

Part 1: Detection of Biomolecules from Solutions with high concentration of Salts and Urea using Probe Electrospray and nano Electrospray

PESI is free from clogging problem and it has high tolerance to salts and urea. We present herein a comparative study of the probe electrospray ionization (PESI) and nano electrospray ionization (nano ESI) for the measurement of biomolecules in the sample solutions with high concentration of salts and urea. Our results show that PESI could provide equivalent ionization performance with nano ESI and in certain cases it could be superior

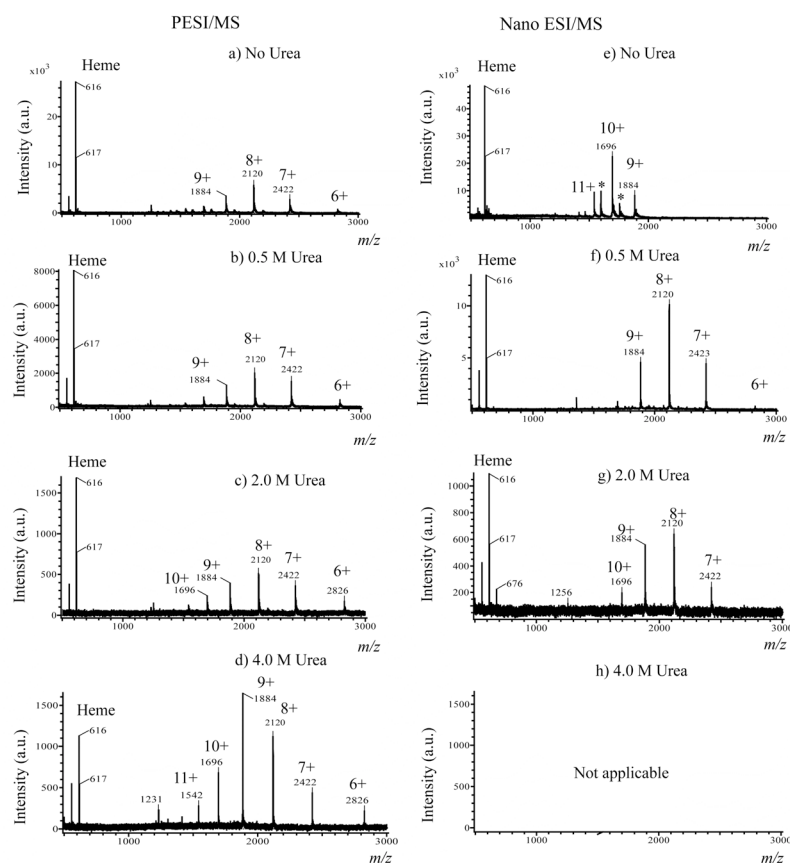


Figure 3. a)-d): Positive ion PESI spectra (using stainless steel acupuncture needle) of 10^{-5} M myoglobin in different concentrations of urea. e)-h): Positive ion nano-ESI mass spectra of 10^{-5} M myoglobin in different concentrations of urea. Peaks labeled with asterisk (*) are those from holo-myoglobin. In h), no mass spectrum could be obtained using nano-ESI-MS due to the severe plugging of the emitter's tip.

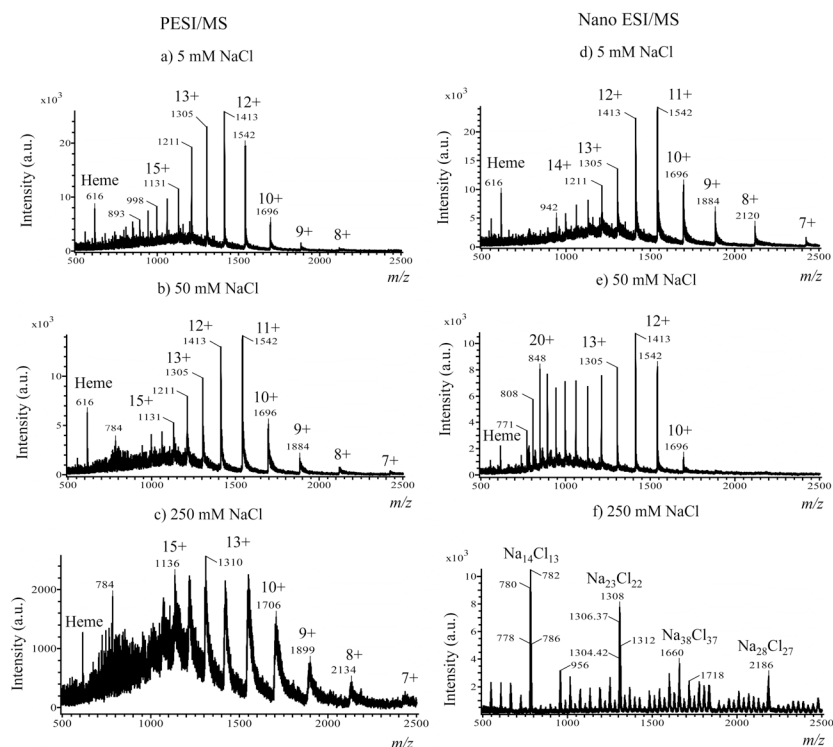


Figure 4. a)-c): Positive ion PESI mass spectra (using titanium wire) for 10^{-5} M myoglobin in different concentrations of NaCl. d)-f): Positive ion nano-ESI mass spectra of 10^{-5} M in different concentrations of NaCl.

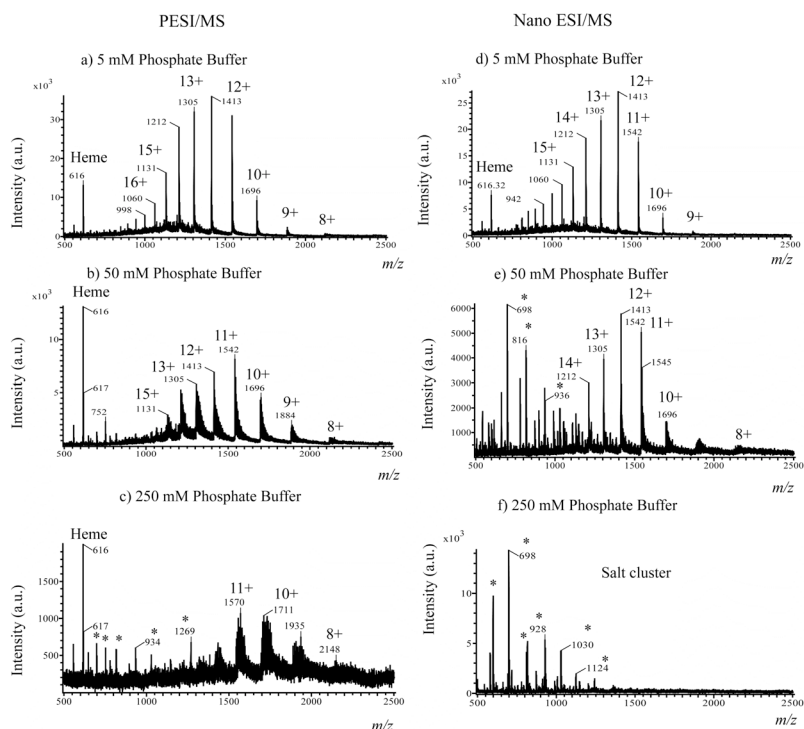


Figure 5. a)-c): Positive ion PESI mass spectra (using titanium wire) of 10^{-5} M myoglobin, in varying concentrations of potassium phosphate buffer. **d)-f):** Positive ion nano-ESI mass spectra of 10^{-5} M myoglobin, in varying concentrations of potassium phosphate buffer (d-f). Peaks labeled with * are originated from the phosphate buffer.

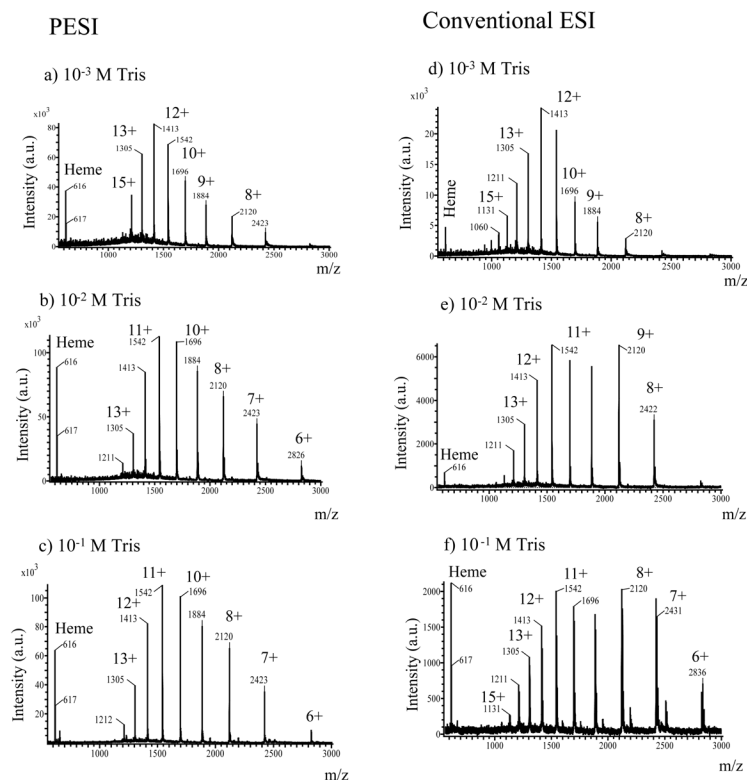


Figure 6. Positive ion PESI mass spectra (using titanium wire) of 5×10^{-5} M myoglobin in 25% MeOH (v/v), 1 % (v/v) HAc and different concentrations of tris (a-c). Positive ion ESI mass spectra of 5×10^{-5} M myoglobin, in 25% MeOH (v/v), 1 % (v/v) HAc and different concentrations of tris (d-f).

to nano ESI for the samples with high concentration (> 100 mM) of salts and urea. Some of the experimental results are shown below.

Part2: Direct Detection of Proteins from Samples with Detergents and Tris Buffer by Single Shot Probe Electrospray Mass Spectrometry (PESI/ MS)

Detergents and tris buffer are exclusively used

for different extraction procedures of biological cell or tissue sample for proteomic research. But they should be separated from sample by other laborious and time consuming methods for further mass spectrometric analysis. Our results show that multiply charged protein ions can be detected directly from the sample solutions with high concentration of different types of detergents and tris buffer using single shot PESI-MS. PESI can selectively detect the protein ions from solutions with 1 M tris, 10^{-2} M of non ionic, 5×10^{-3} M ionic. Some of the preliminary results are illustrated as the followings.

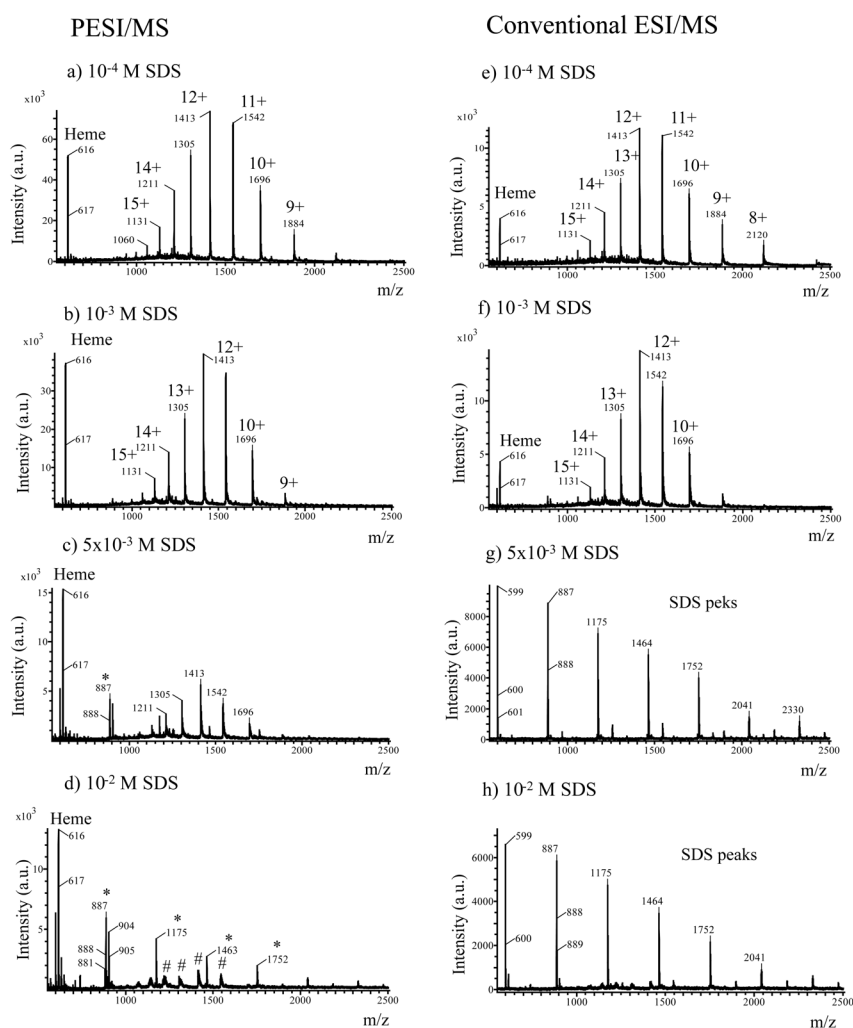


Figure 7. Positive ion PESI mass spectra (using titanium wire) of 5×10^{-5} M myoglobin in 25% (v/v) MeOH, 1 % (v/v) HAc and different concentrations of SDS (a-d). Positive ion ESI mass spectra of 5×10^{-5} M myoglobin, in 25% MeOH (v/v), 1 % (v/v) HAc and different concentrations of SDS (e-h).

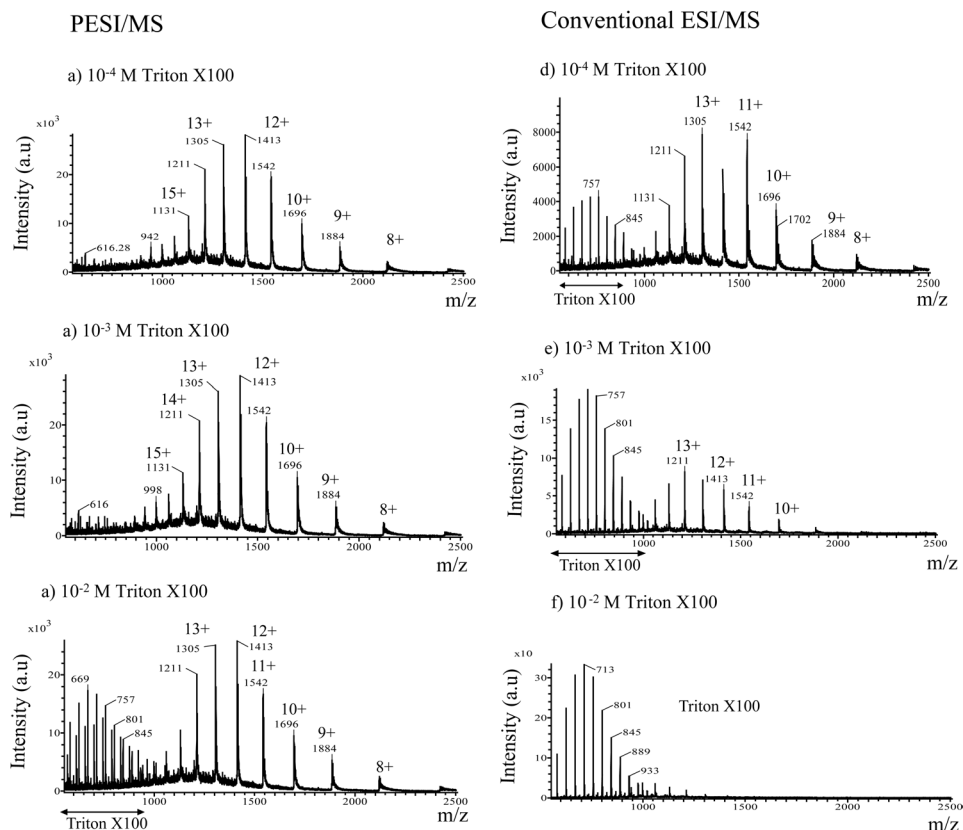


Figure 8. Positive ion PESI mass spectra (using titanium wire) of 5×10^{-5} M myoglobin in 12.5% (v/v) MeOH, 12.5% (v/v) ethyl acetate, 1% (v/v) HAc and different concentrations of triton X100 (a-c). Positive ion ESI mass spectra of 5×10^{-5} M myoglobin, in 12.5% MeOH (v/v), 12.5% (v/v) ethyl acetate, 1 % (v/v) HAc and different concentrations of triton X100 (d-f).

In summary, PESI-MS offers some fundamental properties that favour its use for biomolecules analysis such as its high tolerances towards salt, urea, tris buffers, detergents, the rapid analysis, and the small amount of sample consumption like nano ESI. Optimization with different types of materials for the PESI emitter has also shown to be effective in improving the PESI mass spectra, possibly due to the improved inertness of the needle emitter surface toward the electrochemical process during the electrospray process. Potential applications of PESI include the direct analysis of protein from salts, detergents and urea contaminated solutions, and the high throughput analysis of cell or tissue extraction and digestion of intact tissue etc.

References:

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