

for the proteins. The measurements for the standard ion source were obtained with three different inlet capillaries, all equivalent in dimensions but which differed in performance. For this reason the results for the least sensitive capillary were dropped while the results for the two most sensitive capillaries were averaged, the latter being in good agreement. The RIC ratios derived from the mass spectra are more consistent and are significantly lower than the ratios derived from ion current measured on the octapole given in Table 2. The higher ratios derived from ion current measurements on the octapole can be potentially attributed to a fraction of charged droplets that are carried by vacuum dynamics from the ion funnel to the entrance of the octapole but are unable to travel through the triple quadrupole mass analyzer since the TSQ 7000 employs a non-linear configuration. Furthermore, the standard TSQ 7000 ion source employs an off-axis capillary inlet, i.e. the exit of the capillary is off-axis relative to the entrance of the skimmer cone, which was specifically designed to eliminate solvent spiking of the mass analyzer.

The result in Table 3 for gramicidin S display a gain of 3 times the peak intensity based on its  $2^+$  charge state, the dominant ion in its spectrum under acidic conditions. This observation is in line with the low mass cutoff of the prototype interface i.e. a lower limit in  $m/z$  for which ions are not efficiently transmitted through the device. Work with other singularly charged peptides indicates a nominal cutoff at approximately  $m/z$  500 for the present design and operating conditions. This cutoff and indeed the entire transmission window can be illustrated by comparing the spectrum of polyethylene glycol (average molecular weight, 8000 amu) obtained with both ESI interfaces as shown in FIGS. 9C and 9D. The spectrum taken with the ion funnel yields a transmission window of  $\sim 2$  (i.e. high  $m/z$  low  $m/z$ ) or less than 1000  $m/z$  units at the RF amplitude employed used for these examples.

As expected, the RF amplitude has a direct effect on the  $m/z$  cutoff of the interface and the transmission window. This effect is illustrated with mass spectra obtained using a 29 M solution of horse heart myoglobin as shown in FIG. 10. At first, as the RF amplitude is increased, the signal intensity for all of the charge states (i.e.  $26^+$ – $12^+$ ) increase until the ions of low  $m/z$  (i.e. the high charge states) are unstable by the imposed RF fields and are therefore unable to be transmitted through the ion funnel. Continuing to increase the RF amplitude increasingly shifts the low  $m/z$  cut-off to higher  $m/z$  values. As the low  $m/z$  ions are lost, the higher  $m/z$  ions are more effectively focused through the ion funnel. This effect is shown in FIG. 11 which plots the relative ion current (RIC) and selected peak intensities of individual charge states for the same myoglobin solution. The  $19^+$  charge state ( $m/z$  893.1), typically the base peak in the ESI mass spectrum for denatured myoglobin obtained with a conventional ion source, is the base peak in the spectrum for an RF amplitude of up to  $\sim 100 V_{pp}$  after which its intensity is sharply reduced due to its instability in the higher RF fields. As the RF amplitude is increased the lower charge states (e.g.  $12^+$ ,  $10^+$ , and  $7^+$  shown) sequentially increase in relative abundance. The expected linear relationship is evident by plotting  $m/z$  versus the RF amplitude needed to maximize the peak intensity for a given charge state as shown in FIG. 12A.

Increasing the RF amplitude increased the RIC of the myoglobin spectra to  $150 V_{pp}$  where the overall RIC begins to decline as shown in FIG. 11. Operating the ion funnel at  $150 V_{pp}$ , RF (700 kHz) resulted in an increase in RIC by over 50 times compared to the ion funnel operating in the

DC-only mode. Operation at fixed RF amplitude yielded similar spectra (in terms of ion  $m/z$ ) that increased in signal intensity until about  $70 V_{pp}$  after which the low  $m/z$  cutoff begins to effect the spectrum by progressively removing the highest charge state on the lower  $m/z$  end of the spectrum. Since the effect of RF amplitude on the low  $m/z$  cutoff is linear with  $m/z$ , this bias can be used to reduce space charge limits (and improve ion focusing through a conductance aperture) and/or remove low  $m/z$  species from contributing to the capacity of ion trapping instruments.

As shown in FIG. 10, at RF levels above  $100 V_{pp}$ , there are a multitude of peaks that appear in the region of the low  $m/z$  cut-off. These are products of collisional induced dissociation (CID) and originate from increased translational energy of low  $m/z$  ions near their stability limit in the ion funnel at the given RF amplitude. Contributions from CID can be effectively minimized by scanning the RF amplitude in-link with the  $m/z$  scan of the quadrupole mass analyzer. This method of scanning would also bring in the maximum intensity for all of the charge states produced by the ESI process. This advantage is illustrated by plotting the maximum peak intensities of the given myoglobin charge states and comparing them to the charge state intensities obtained with most sensitive capillary inlet used on the standard ESI ion source as shown in FIG. 12B. A secondary benefit is that moderate amounts of collisional activation can be produced in the ion funnel to reduce contributions due to charge state adduction. Note that in FIG. 10 adducts associated with lower charge states are reduced as the RF level is increased.

While a preferred embodiment of the present invention has been shown and described, it will be apparent to those skilled in the art that many variations, changes and modifications may be made without departing from the invention in its broader aspects. The appended claims are therefore intended to cover all such changes and modifications as fall within the true spirit and scope of the invention.

We claim:

1. A method of focusing dispersed charged particles comprising the steps of:

- providing a plurality of elements in a region maintained at a pressure between  $10^{-1}$  millibar and 1 bar, each of said elements having successively larger apertures wherein said apertures form an ion funnel having an entry at the largest aperture and an exit at the smallest aperture,
- applying an RF voltage to each of the elements wherein the RF voltage applied to each element is out of phase with the RF voltage applied to the adjacent element(s),
- directing charged particles into the entry and out of the exit of the ion funnel, thereby focusing the charged particles.

2. The method of claim 1 further comprising the step of directing the charged particles is provided by a mechanical means.

3. The method of claim 2 wherein the mechanical means is selected from the group comprising a fan, a vacuum, or combinations thereof.

4. The method of claim 1 further comprising the step of directing the charged particles by providing a DC potential gradient across the plurality of elements.

5. The method of claim 1 further comprising the step of directing charged particles generated in a multi-inlet system into the ion funnel.

6. The method of claim 1 further comprising the step of providing a plurality of said ion funnels in series.

7. The method of claim 1 wherein the exit of said ion funnel is provided adjacent to a multipole lens element.