**Digital Waveform Technology and Ultra High Mass Spectrometry**

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**Introduction**

Digitally driven quadruple mass filters, ion guides, and traps are the future of mass spectrometry. Simply by manipulating the duty cycle and frequency of the applied waveforms, novel and useful abilities have been demonstrated that can be used for the analysis of large ions in low charge states. This technology is game changing because it affects so many areas of mass spectrometry. The goal of this poster is to reveal some of these advances.

**High Resolution Digital Quadrupole Time-of-Flight Mass Spectrometry**

The duty cycle of the applied rectangular waveforms (defined by the fraction of time the waveform is in the high state) can be manipulated to allow an ordinary ion guide to become a sophisticated linear ion trap that can axially collect and eject ions on demand (see Fig. 1).

Fig. 2 reveals the Faraday detector response that shows more than 1.5 million intact singly charged insulin ions, can be axially ejected in a temporarily short (200 μs) focused ion plug into an awaiting orthogonal time-of-flight mass analyzer. Spatial focusing is not mass dependent; therefore, resolving power does not change with m/z.

**Digital Mass Filter Analysis, Stability Diagrams, and Pseudopotential Well Depth**

In Fig. 4 the Mathieu stability diagrams for sine (a), square (c), and rectangular (e) waves are depicted. Within the red outlines, the ions are stable. The gray scale in the diagram defines the pseudopotential well depth. Darker shades reveal deeper wells that defines the strength of binding of the ions in the ion guide. The cyan lines in the diagrams depict the scan lines for mass filter analysis. (b), (d), and (f) show close-ups of the apices, where the region along the cyan line creates a narrow range of stable values of m/z.

Our group pioneered the method of defining well depth at all points in Mathieu (q, a) space and translating it into the laboratory frame so that different methods of mass filter analysis can be compared. Fig. 5 compares a snapshot of the well during a mass scan for typical sine (a) and rectangular (b) wave operation at unit resolution (Am = 1) and m/z 1000. The shapes of the well correlates with the shape of the mass peak, while the well depth defines the ability of the mass filter to transmit ions, deeper wells—better transmission/sensitivity. We can conclude that typical sine wave operation yields better transmission/sensitivity. The outcome switches when the mass filters are operated in higher stability zones. Fig. 6 and 7 reveal the m/z 1000 well at 700 V for 62.5/37.5 and 70.0/30.0 duty cycle waveforms, respectively. The baseline resolving powers are approximately 1300 and 36,000 with well depths of 11 and 7 V, respectively. These higher stability zones represent orders of magnitude increases in resolving power and sensitivity.

Because well depth is proportional to operating voltage, calculations in MHz waves are made by finite integers. 10 ppm clock (see Fig. 10). Duty cycle control needed for filter analysis. Still can still do a lot with DWT and edge counting. For example axial trapping and ejection shown in Figs. 1-3.

**Digital Waveform Generation**

In order to realize digital waveform-based mass filter analysis in any stability zone, waveforms must be generated with high resolution duty cycles.

**The old way**

Previously DWT could not produce a credible mass filter—not enough duty cycle control. Rectangle waveforms were made by counting edges of a master clock (see Fig. 10). Duty cycle resolution was limited by finite integers. 10 ppm control needed for filter analysis. Still can still do a lot with DWT and edge counting. For example axial trapping and ejection shown in Figs. 1-3.

**The new way**

Direct Digital Synthesis is used to make smooth sine waves with pH2 resolution on MHz waves (see Fig. 3). Our method compares sine wave output voltage to a DC level set by a high res 18-bit DAC. When the sine wave potential is greater than the DAC generated comparator voltage, the comparator output is high and low otherwise. Our method provides 10 ppm duty cycle resolution. That is enough to create a mass filter without using a DC potential (a=0). The waveform generator was created (see Fig. 12).

A mass filter spectrum lysozyme and its multimers demonstrating the 1st stability zone analysis is shown in Fig. 13. Fig. 14 shows a stability zone 3 mass spectrum of lysozyme with a 76.6/23.4 duty cycle.

**New Methods of Performing MSn in Linear Ion Guides**

**Boundary and Frequency Hopping Induced Excitation**

In Fig. 15 (a) and (b) reveal the stability diagram change with duty cycle. Ions are finite isolated and then trapped at the point illustrated in (a). Then the stability boundary is jumped by switching the duty cycle to induce excitation for 5 cycles and jumped back to relax for 50 cycles. The excitation/relaxation process was repeated 10 times (c) and (d) reveal the mass spectra before and after boundary jumping induced excitation for 5 cycles and relaxation for 50 cycles.

Fig. 16 (a) and (b) depict the duty cycle induced changes to the stability diagrams while (c) and (d) show the before and after effect of parking the ions just inside the boundary for 100 ms (50,000 cycles). The stability boundary for a specific value of m/z defines the point of maximum translational energy while remaining trapped. Parking the ions at the boundary for an extended period slowly increases the ion’s internal energy for fragmentation.

**Future Directions**

Our group is beginning to experimentally explore mass filter analysis in higher stability zones. We plan to apply this to mass analysis of intact proteins and complexes in low charge states. The higher stability zone mass filter analysis will be leveraged to produce miniaturized low voltage mass filter instruments that can be used for MS/MS analysis. We also intend to explore the production of stable isotopes using digital mass filters.

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Fig. 1. Fig. 2. Peak center assignment is countaus. Fig. 3. 1.6 ppm, expect 100 ppm with a good mass filter. Fig. 4. Fig. 5. Fig. 6. Fig. 7. Fig. 8. Fig. 9. Fig. 10. Fig. 11. Fig. 12. Fig. 13. Fig. 14. Fig. 15. Fig. 16.