

Adding MS/MS Capability to a Single Quadrupole Mass Spectrometer.

Waters™

Martin Green*, David Douce, Thomas Moore, Witold Niklewski, Ashley Sage.
Waters Corporation, Wilmslow, UK

OVERVIEW

PURPOSE:

Development of a prototype Single Quadrupole / Linear ion trap (LIT) hybrid instrument by adding MS/MS capability to a single quadrupole.

METHOD:

Modifications to Waters QDa™ Mass Detector with no change in mass-filter functionality or instrument footprint. Precursor isolation during trap fill using pulsed DC.

RESULTS:

MSⁿ and MRM-like (Multiple Reaction Monitoring) targeted modes were demonstrated using both in-trap resonance dissociation and beam-type CID in up-stream source ion guide.

INTRODUCTION

Commercial single quadrupole mass spectrometer (SQMS) functionality (Full scan MS and Single Ion Recording (SIR)) has remained essentially unchanged since first commercialized in 1964. In this presentation we describe adding MS/MS functionality to a SQMS with no change to basic functionality or footprint. This is realized by operating the quadrupole as a mass-filter or as a linear ion trap (LIT) with mass selective axial ejection. Addition of MS/MS greatly increases the specificity of the SQMS allowing enhanced compound characterization, and targeted analysis from complex mixtures.

METHODS

A Waters QDa™ Mass Detector was modified to allow ions to be trapped, isolated, dissociated and mass selectively ejected without compromise to mass-filtering functionality. Figure 1.

- Addition of Short 'post-filter' rods to allow trapping and mass-selective axial ejection.
- Modifications to apply dipolar excitation and to allow the pressure in the quadrupole chamber to be adjusted.
- Control software and electronics modifications to allow complex ion-manipulation sequences during inter-scan periods.

All data were recorded directly into Waters MassLynx™ Software for post-processing.

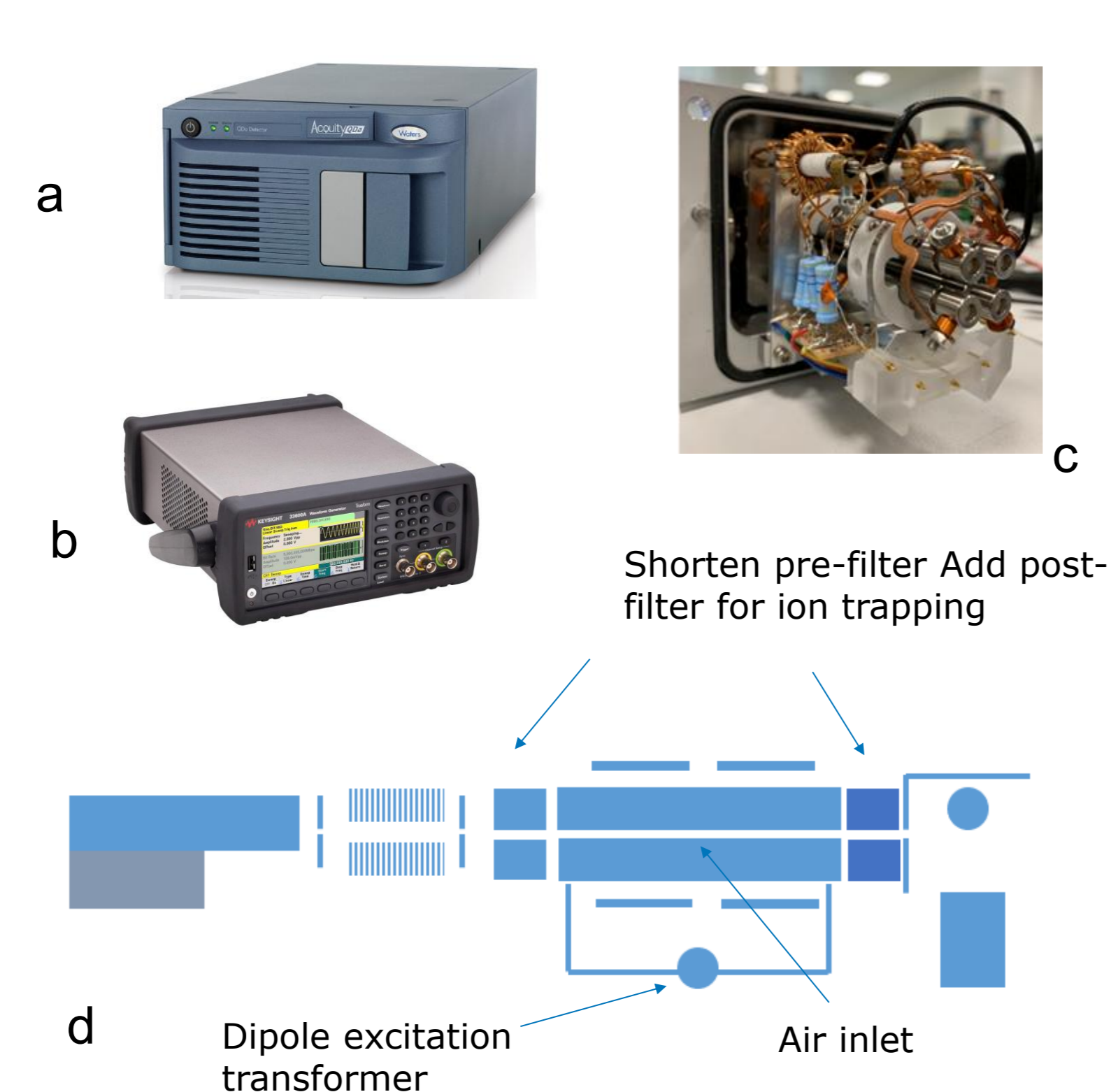


Figure 1 a. Waters QDa Detector b. Keysight 33622A waveform generator for dipole excitation. c. Modified hardware, d. schematic showing modifications.

PRECURSOR ISOLATION

Avoiding space-charge distortion requires precursor isolation during the trap 'fill-time'. A combination of static resolving DC (~100 amu window) and pulsed resolving DC (~5 amu window) was used. Figure 4. To trap and cool ions, analyser pressure was raised to ~1 x 10⁻⁴ Torr of air. With resolving DC applied, stability depends strongly on time and pressure. Figure 2, Figure 3.

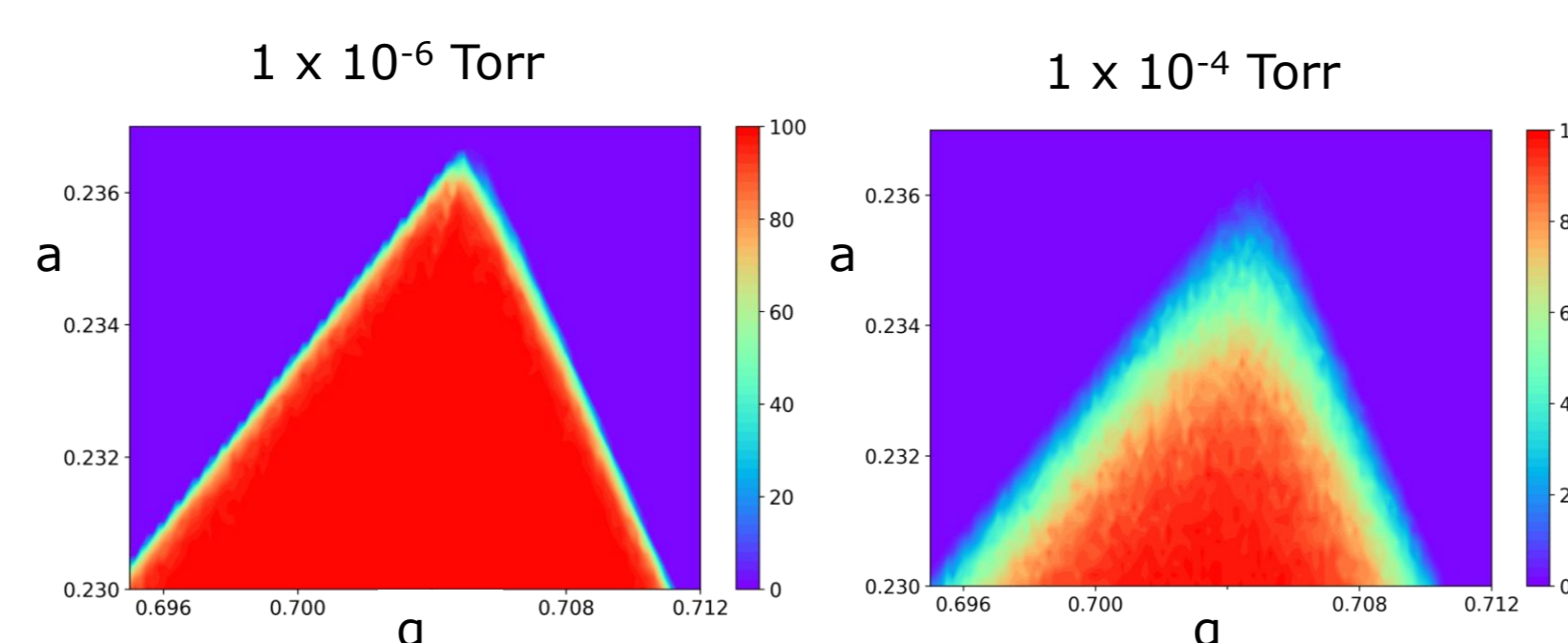


Figure 2 Mathieu Stability Diagrams (SIMION™) for 1 ms trapping m/z = 500, round quadrupole rods, r0 = 3.55 mm. A 1 x 10⁻⁶ Torr N₂. B 1 x 10⁻⁴ Torr N₂

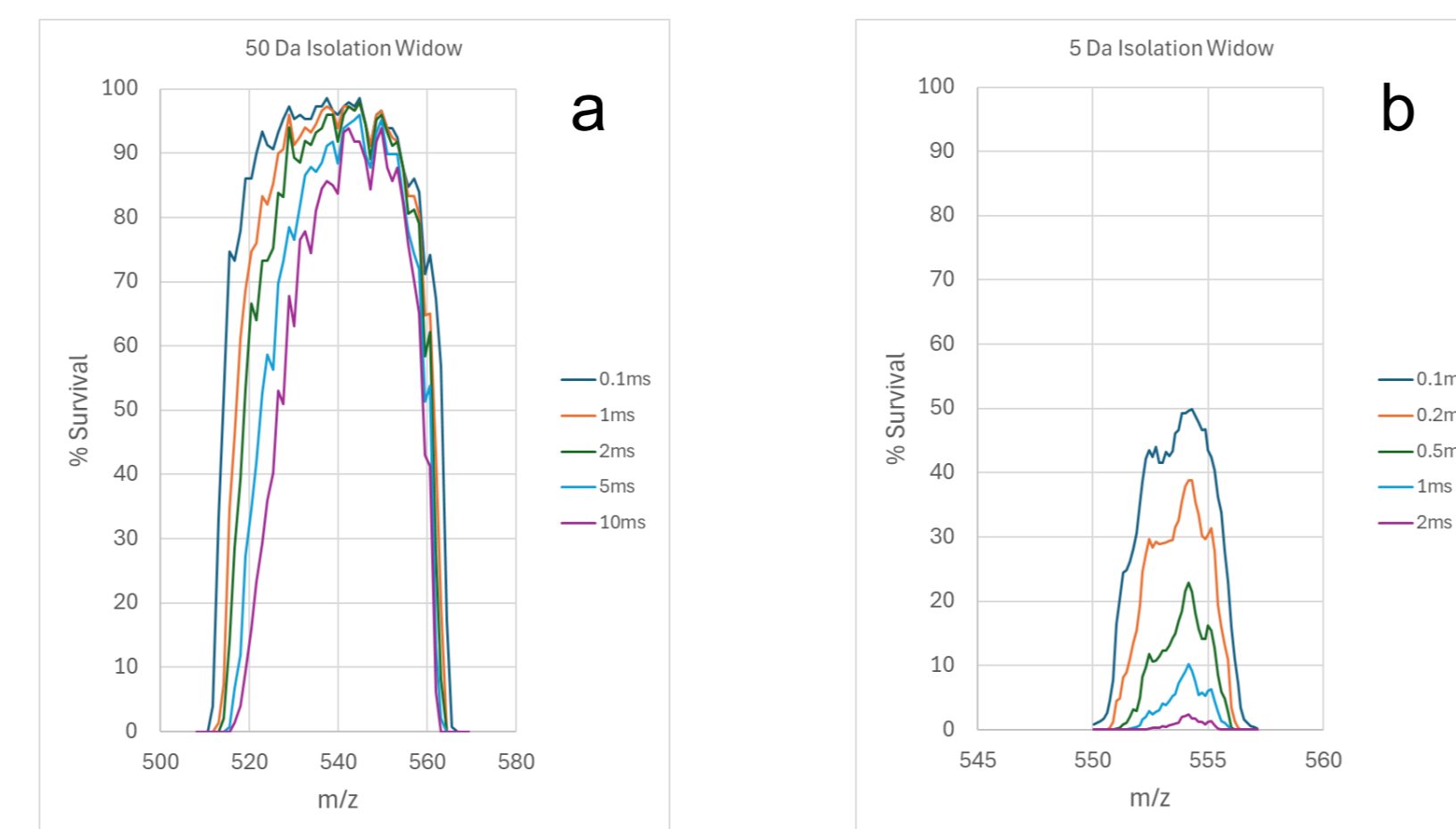


Figure 3 Ion survival (%) in LIT vs m/z, w.r.t trapping time (ms). 1 x 10⁻⁴ Torr N₂. a. 50 amu isolation window 0.1 ms - 10 ms. b. 5 amu isolation window 0.1 ms - 2 ms.

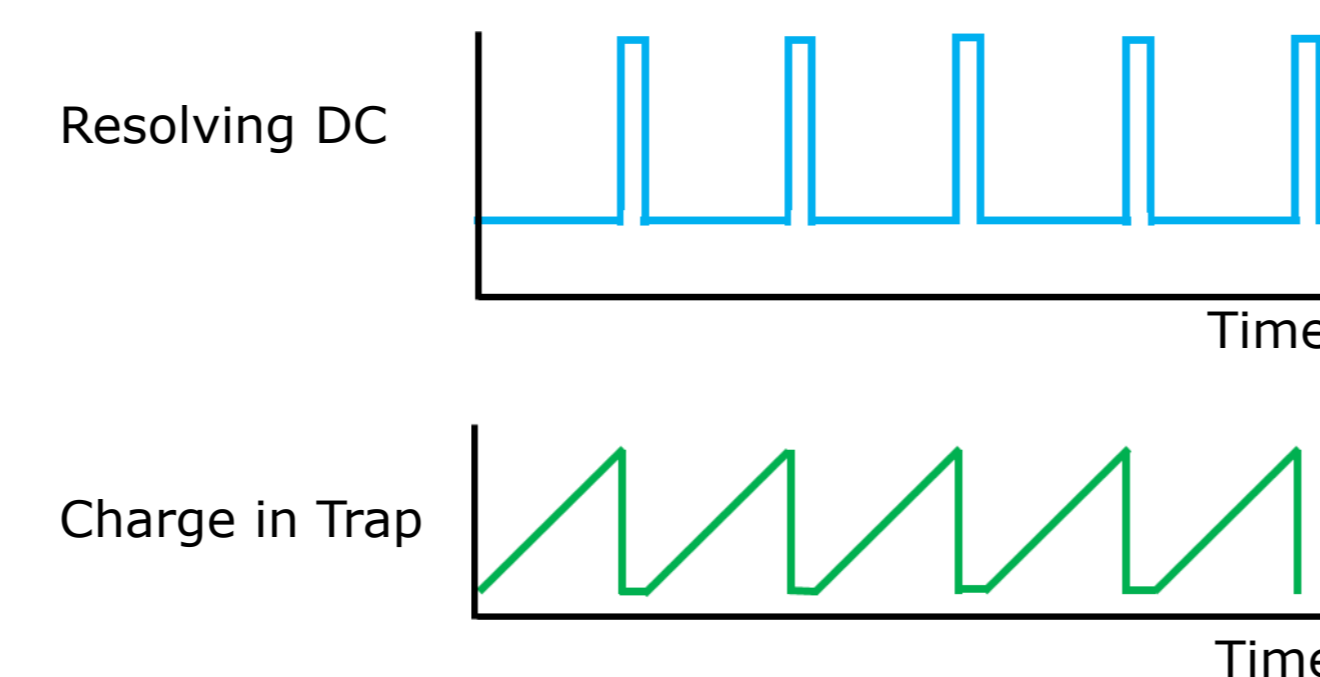


Figure 4 Operation of pulsed resolving DC during fill-time to control maximum charge in the LIT.

MODES OF OPERATION

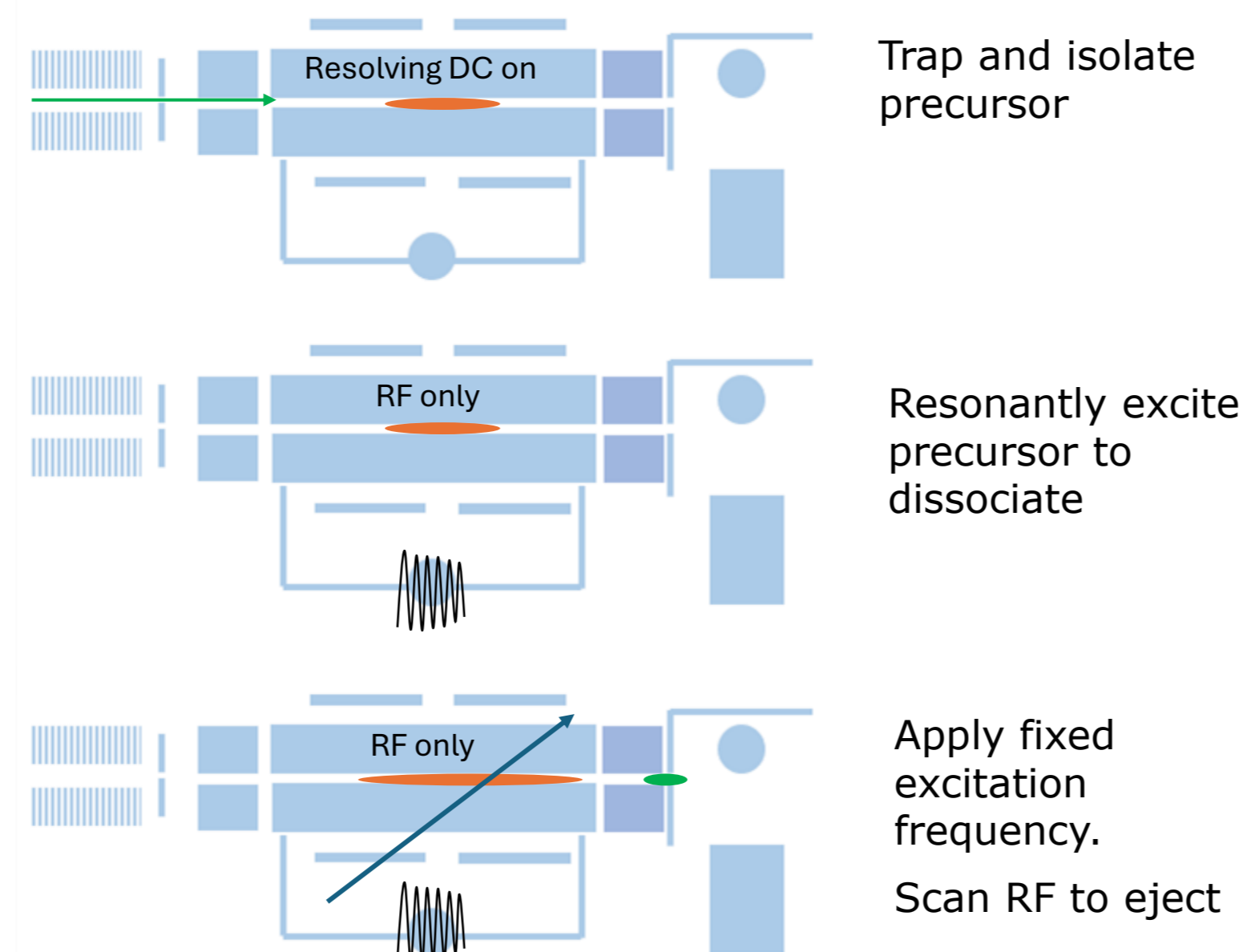


Figure 5 Full scan MS/MS mode with resonance dissociation

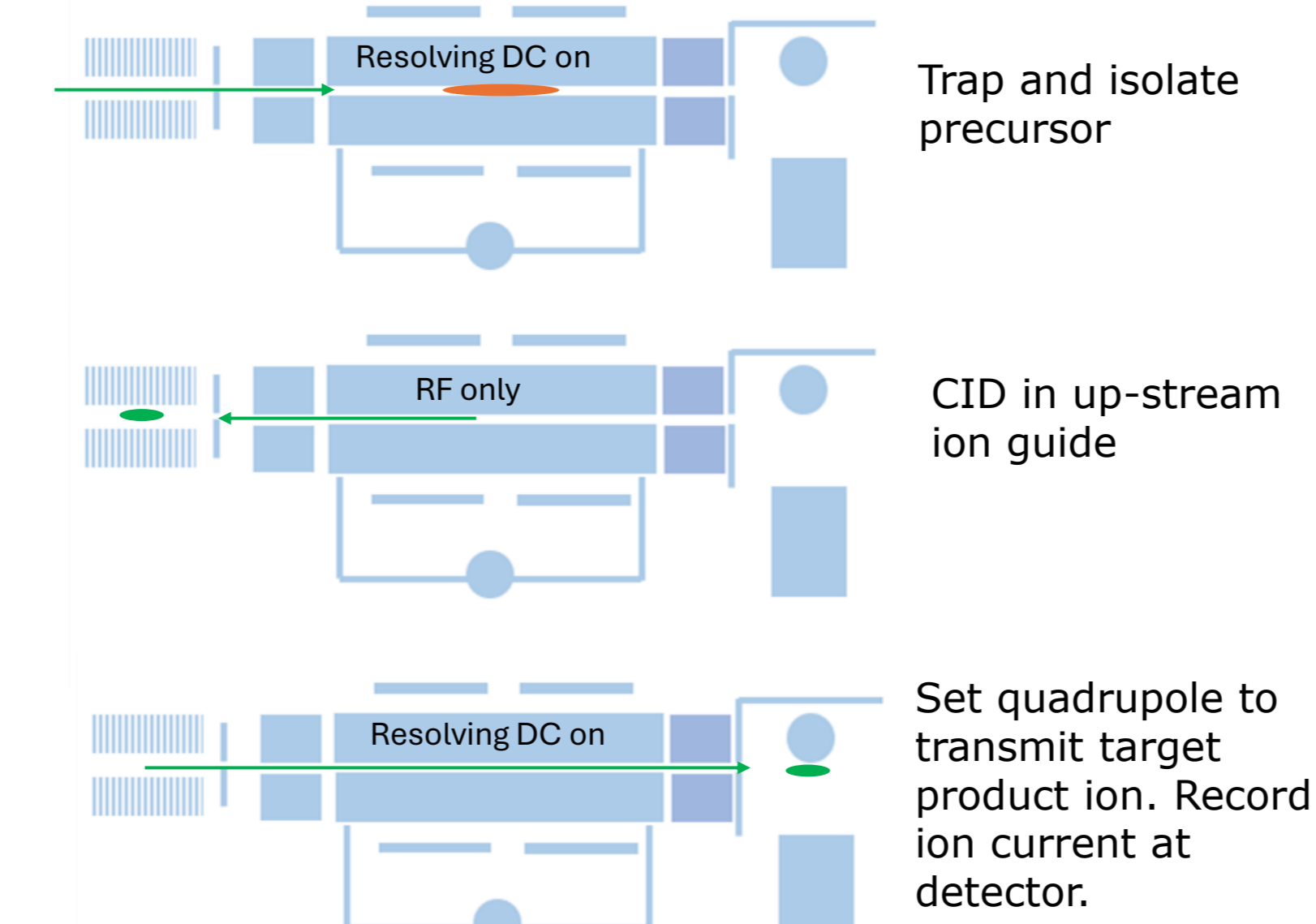


Figure 6 Targeted MRM-like mode with CID.

Figure 5 and 6 illustrate two modes of operation, full scan and targeted MS/MS. Both resonance dissociation and beam-type CID are accessible as well as MSⁿ operation and other targeted modes using resonance ejection of one or more product ions.

RESULTS

Full scan MS/MS resonance dissociation.

Figure 7. Shows the full scan MS/MS spectrum of Angiotensin I, [M+3H]³⁺ m/z = 432.9, 850 amu/sec analytical scan, 16 ms excitation at 100 mV pk-pk.

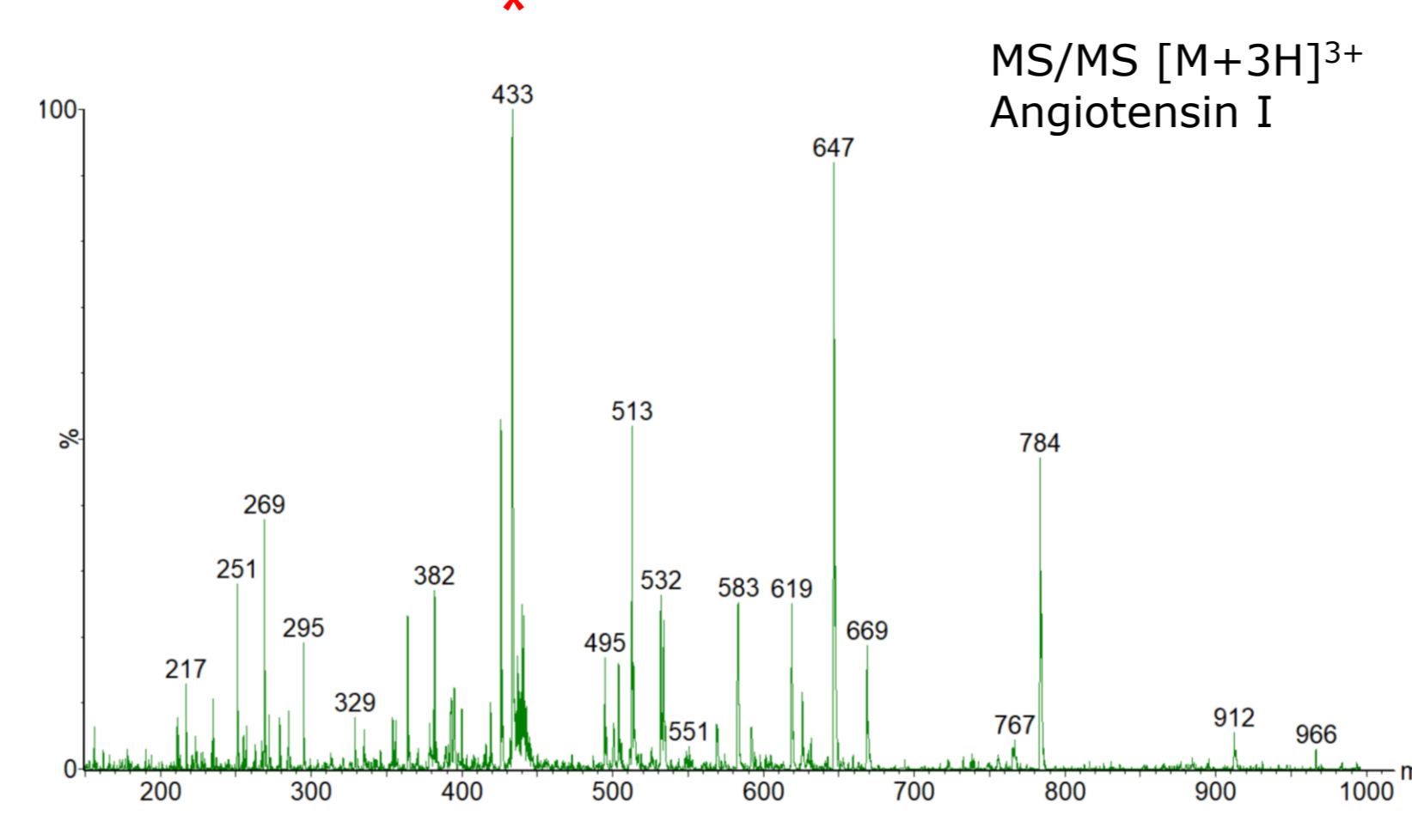


Figure 7. Full scan MS/MS Angiotensin I, [M+3H]³⁺ m/z = 432.9

MS/MS CID vs resonance dissociation

Figure 8 shows a comparison of MS/MS of [M+H]⁺ Leucine Enkephalin m/z 556.3 using CID (30 eV) in the source ion guide and resonance dissociation (12 ms, 100 mV) in the trap.

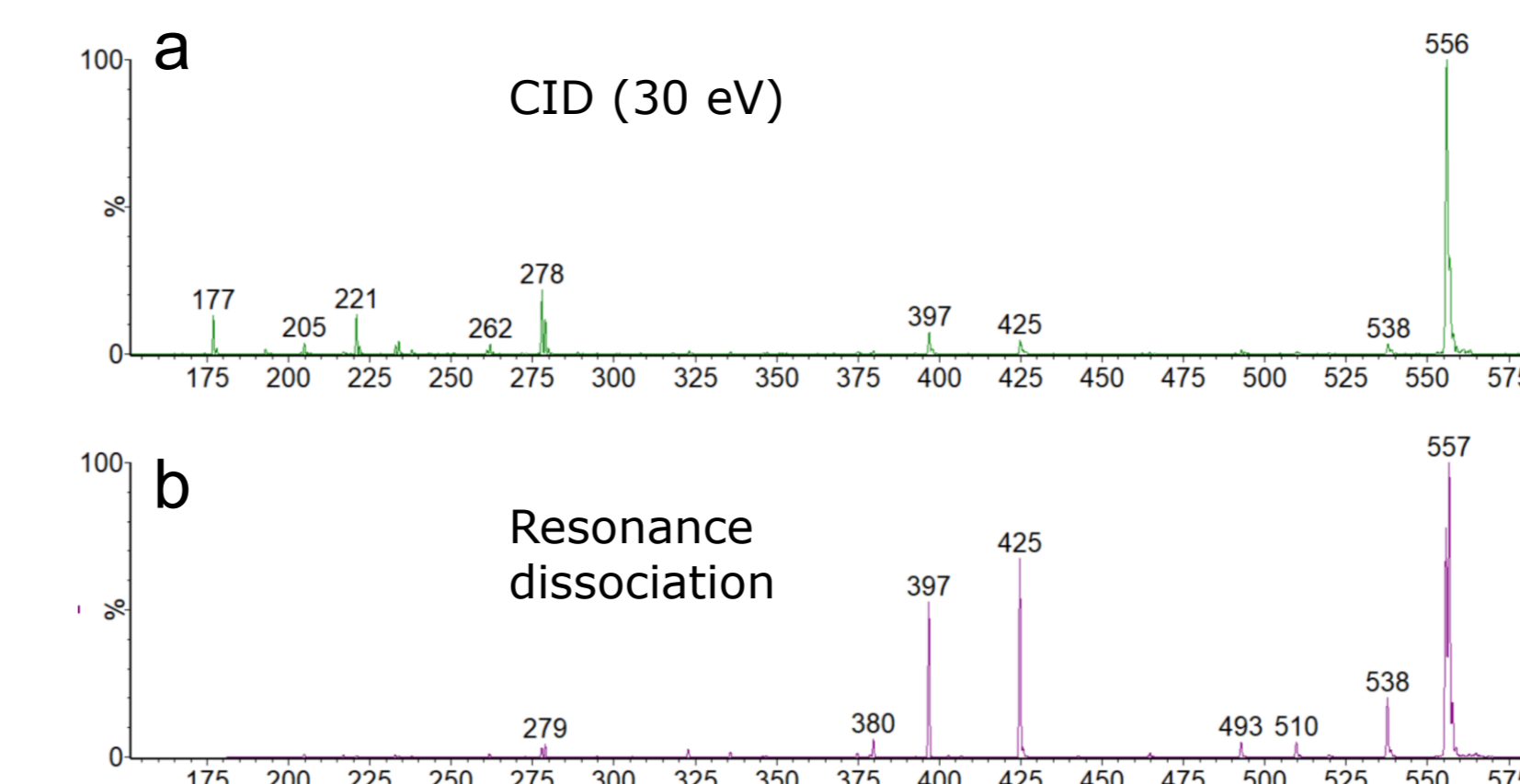


Figure 8. Full scan MS/MS (2000 amu/sec) Leucine Enkephalin m/z 556.3 a. CID, b. resonance dissociation

MS³ Leucine Enkephalin

Figure 9 shows MS/MS [M+H]⁺ Leucine Enkephalin m/z 556.3, followed by MS³ of product ion at m/z = 397.2

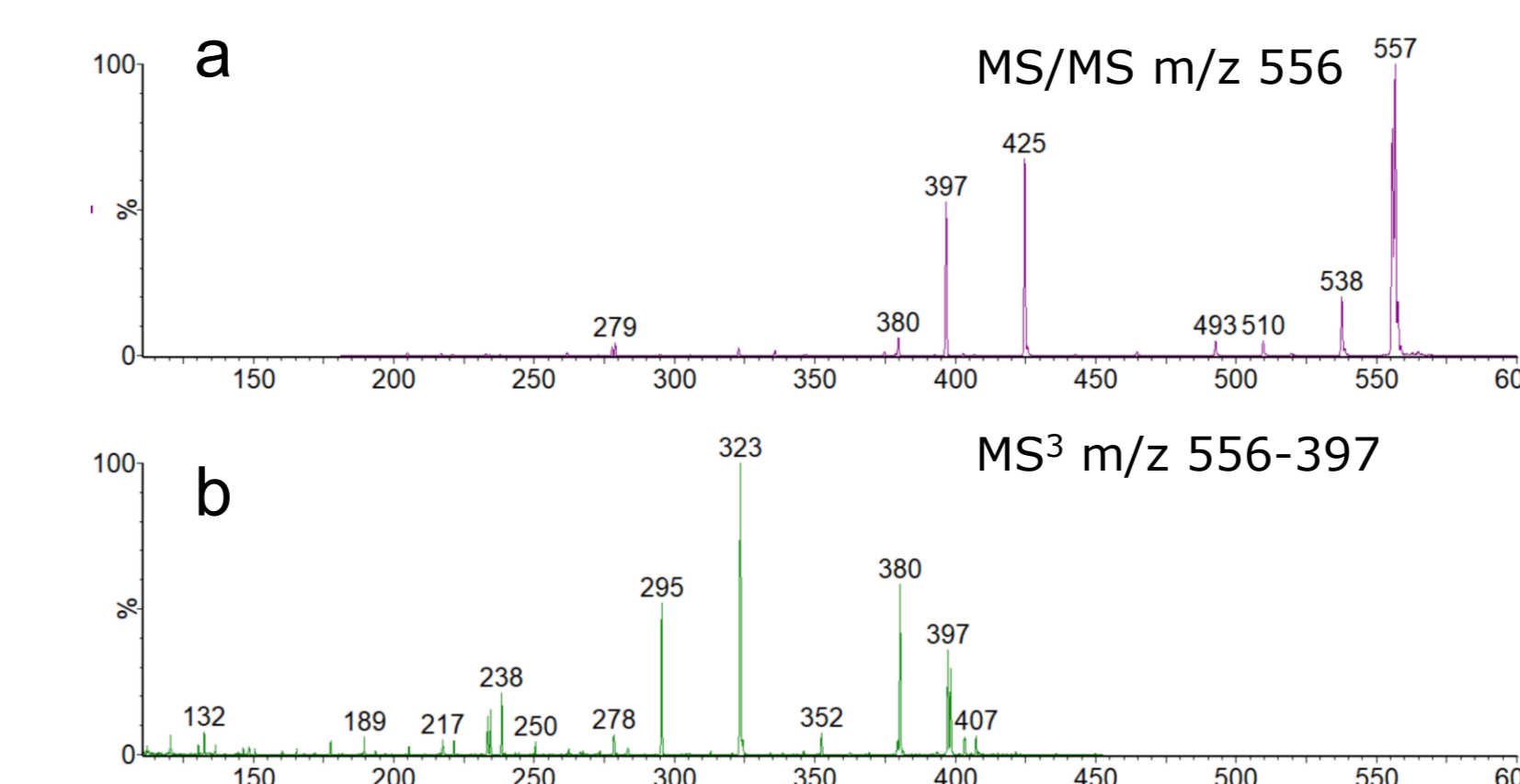


Figure 9 a. Full scan MS/MS (2000 amu/sec) Leucine Enkephalin m/z 556, b. MS³ of product ion at m/z 397

LC-MRM mode with CID (19 eV)

Figure 10. shows LC-MRM chromatograms from the transition m/z 321-152 for a series dilution of Chloramphenicol in negative ion. The total cycle time for precursor isolation and trapping; CID in the up-stream ion guide; setting the quadrupole in mass-filter mode (m/z 152); ejection and detection was 25 ms.

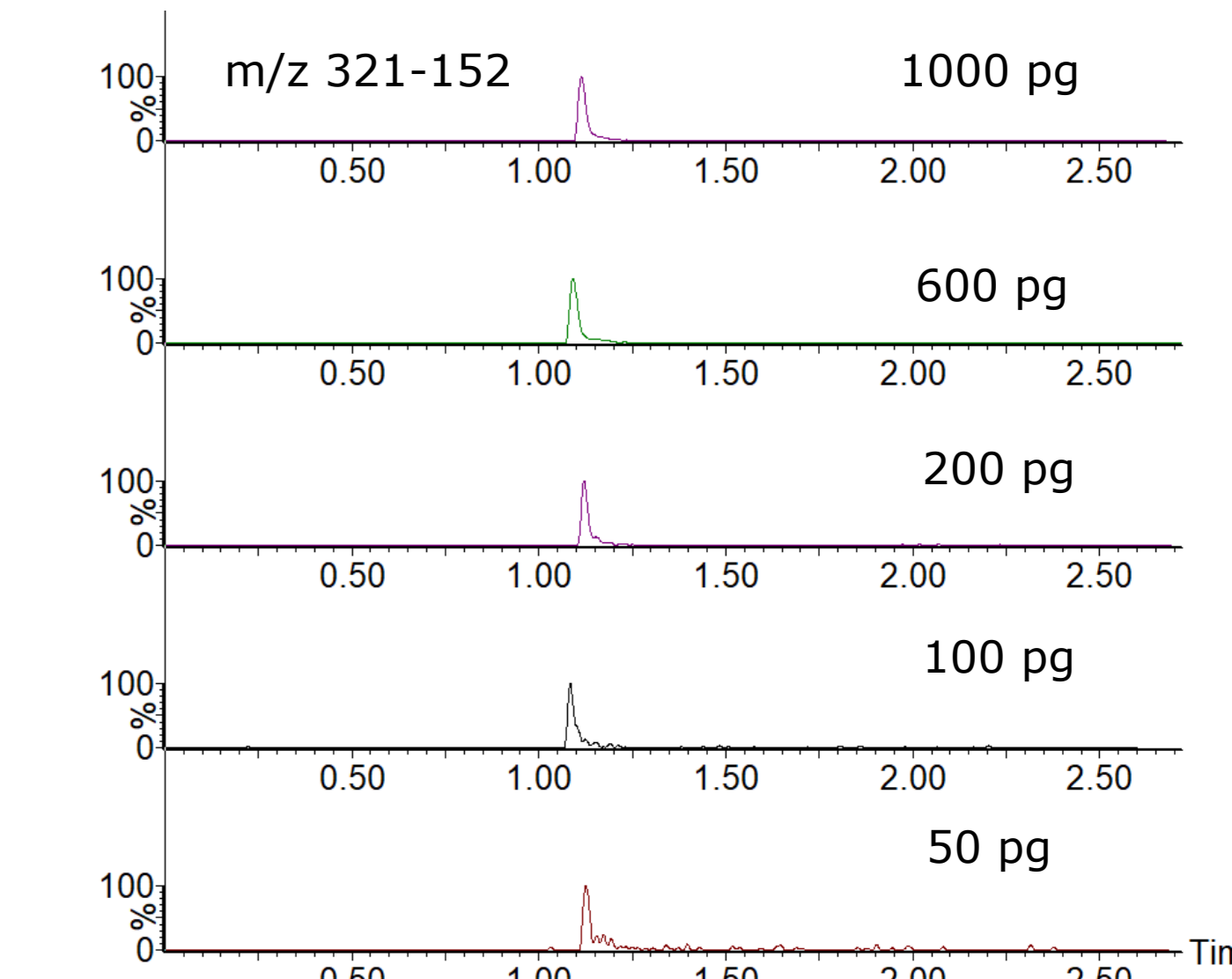


Figure 10. LC-MRM chromatograms for a serial dilution (50 pg - 1000 pg on column) of Chloramphenicol, negative ion mode.

DISCUSSION

This work demonstrates that, with simple modifications and no loss in SQMS performance, powerful linear ion trap functionality can be realized. This transforms the SQMS system into a tool for deep structural characterization, with different dissociation schemes, MSⁿ operation, and tandem quadrupole type specificity. Slower LIT scan speeds allow access to enhanced mass resolution to distinguish higher charge states. Many other operational modes can be envisaged expanding SQMS applications into other areas.

CONCLUSION

The prototype SQMS/LIT instrument is capable of high-quality MS/MS operation with simple modifications, no loss in mass-filter performance and no change in instrument footprint. This represents the first demonstration of true MS/MS and MSⁿ capability on a compact commercial SQMS platform.

Both CID and resonance dissociation are available along with targeted MRM-like modes, all compatible with UHPLC timescales. Future work includes, optimising duty cycle, high resolution zoom scans, and integration with ambient ionization techniques such as ASAP.

The authors are employees of Waters Corporation, and this work is presented on behalf of the company.

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