The Power of High Resolution Accurate Mass Using Orbitrap Based GC-MS

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Key Words

Gas Chromatography, High Resolution, Mass Accuracy, Orbitrap Technology

Introduction

The Thermo Scientific™ Q Exactive™ GC hybrid quadrupole-Orbitrap mass spectrometer is a benchtop instrument, designed to bring the power of Orbitrap high-resolution, accurate-mass (HR/AM) to gas chromatographic (GC) separations.

Obtaining mass spectrometer (MS) measurements with high mass accuracy is essential to providing the required selectivity in complex matrices and to increasing the confidence in compound identification and confirmation. For the former, obtaining a consistently high mass accuracy allows the use of very narrow mass extraction windows, taking full advantage of the instrument's mass resolving power. For the latter, measuring the mass of a chemical with sufficient accuracy allows the chemist to predict the elemental composition and isotopic ratios to help identify the chemical structure of the substance.

The results of this work demonstrate that, using the high resolution of the Q Exactive GC mass spectrometer, excellent mass accuracy is always available to the user, across a wide concentration range and in the presence of a complex chemical background.

Experimental

The Q Exactive GC system was tuned and calibrated using peaks of known mass from a calibration solution (FC 43, CAS 311-89-7) to achieve mass accuracy of < 0.5 ppm RMS. As illustrated in Figure 1, the system was operated in electron ionization (EI) mode using full scan and various resolving powers (full width at half maximum or FWHM). Data was lock-mass corrected during the acquisition using GC column bleed siloxane masses.

Table 1. Mass spectrometer conditions.

Mass Spectrometer Parameters	
Transfer line (°C):	280
Ionization type:	EI
Ion source (°C):	230
Electron energy (eV):	70
Acquisition Mode:	full scan
Mass range (Da):	50–500
Mass resolution (FWHM at m/z 200):	15k, 30k, 60k, and 120k
Lockmass, column bleed (m/z):	207.03235

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## Selectivity Through Accurate Mass

With Q Exactive GC technology, one can achieve selective detection of target analytes in complex matrices through the use of very high resolving power that delivers sub-ppm mass accuracy. This capability is demonstrated in Figure 1, where a leek sample extract (spiked with several pesticides at the 10 ng/g level) was acquired in full scan at 60k resolving power (FWHM at *m/z* 200). Extracting the exact mass for iprodione (*m/z* 314.0094) with a ±5 ppm mass tolerance window enables generation of a highly selective extracted ion chromatogram (XIC) with reduced chemical interferences from the leek matrix background. In contrast, a nominal mass extraction window (±3,184 ppm), simulating unit-mass resolution acquisition, will not provide enough selectivity to detect this pesticide (Figure 1).

Figure 1. Full scan accurate mass selectivity demonstrated for iprodione at 10 ng/g in a leek sample. Accurate mass measurements enable confident detection (±5 ppm) (top), whereas at nominal mass acquisitions this pesticide is not detected (±3,184 ppm) (bottom).
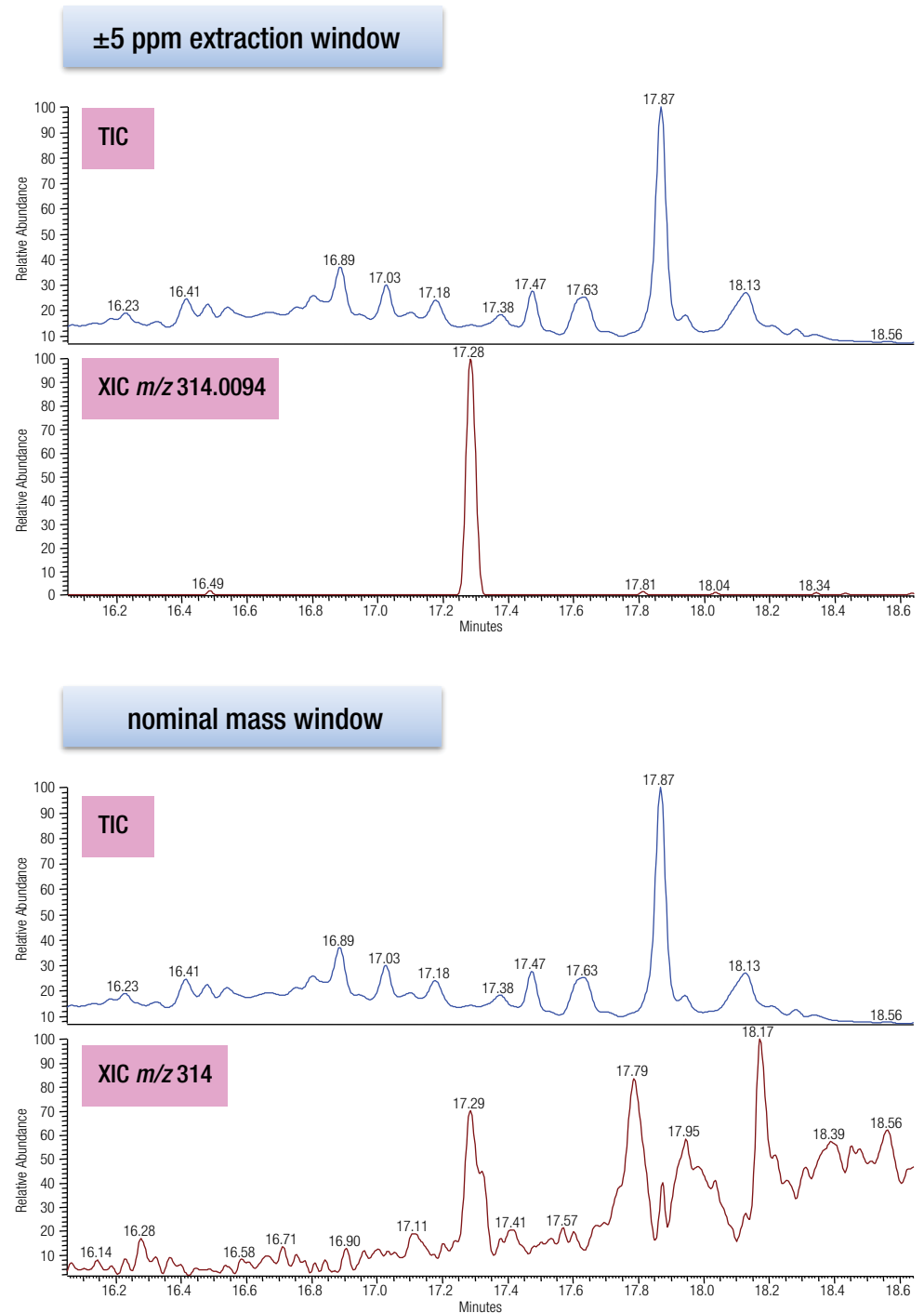
Obtaining accurate and consistent mass measurements increases the confidence in compound identification and elemental composition assignment. The number of possible chemical formulae that can be assigned to an unknown compound can be significantly reduced if the accuracy of a measured mass is high. The relationship between accurate mass measurements and the number of possible chemical formulae is shown in Figure 2, where a 50 ppm mass accuracy window generates (from the pre-selected number of C,H, O, N and Cl) 147 possible chemical formulae that correspond to the measured mass *m/z* 314.00940.

In contrast, a <1 ppm mass accuracy window suggests only two possible elemental compositions assigned to this mass. In order to further discriminate between these two chemical formulae one can compare the theoretical isotopic pattern of each proposed chemical formula with the measured isotopic pattern. As shown in Figure 2, it is then obvious that the resolving power and mass accuracy are intrinsically linked and using enough resolution to separate the target ions from the background chemical interferences is very important for accurately determining and confirming the mass of that target chemical.

Figure 2. The effect of mass accuracy on the number of possible chemical formulae (generated from the selected chemical elements shown in the inserted table) for a measured mass of *m/z* 314.00940 (measured at 60k FWHM resolution). Increased number of possible formulae (annotated on top of the bars) with increase mass uncertainty. Even 1 ppm mass accuracy can lead to two possible chemical formulae assigned to the measured mass and, in this case, isotopic pattern comparison (measured versus theoretical) can be used to eliminate possible chemical formulae (bottom left) and confirm the most probable chemical composition (bottom right).
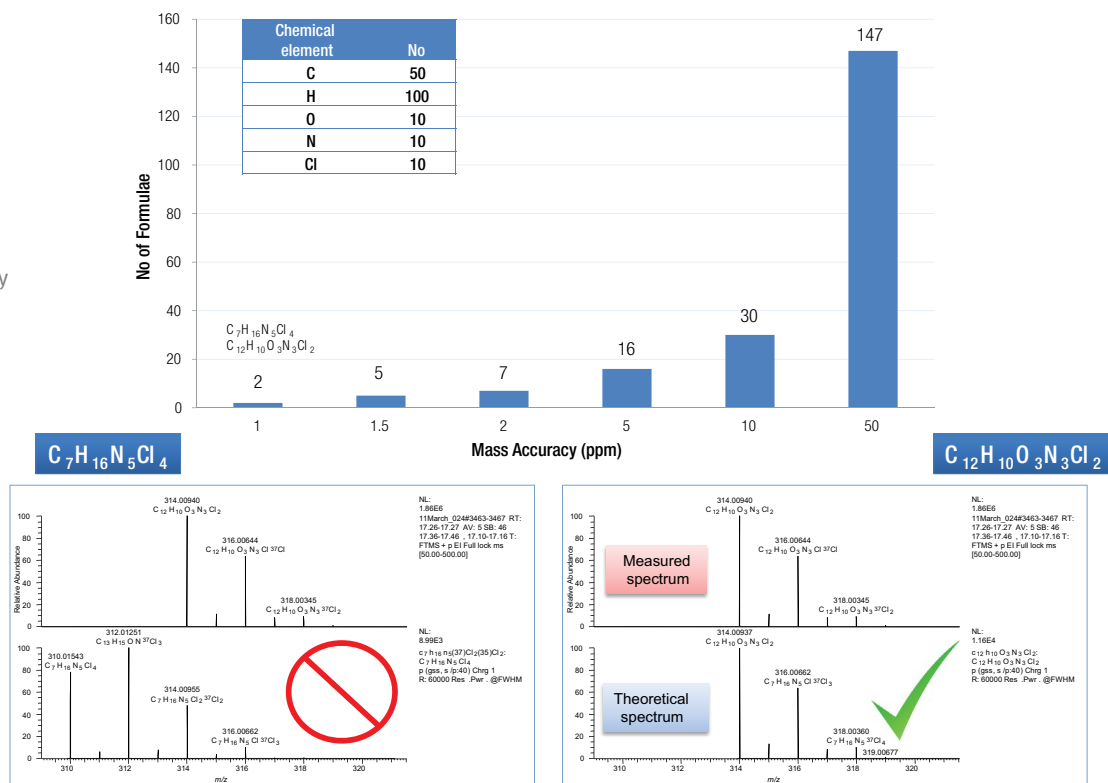
In the experiments conducted, it was observed that for resolving powers of 60,000 FWHM (*m/z* 200) and higher, mass differences of several pesticides are less than 1 ppm. In contrast, for resolving powers around and below 30,000 FWHM (*m/z* 200) higher mass errors are measured. These higher mass errors are due to inference of chemical background ions with the target ions (Figure 3).

Figure 3. The effect of resolving power (reported at *m/z* 200) on mass accuracy measurements of chlorpropham, procymidone, and iprodione in a leek sample spiked with pesticides at 10 ng/g. A resolving power higher than 30k is required to completely separate these pesticides from the interfering matrix ions and to deliver <1 ppm mass accuracy.

	120k	60k	30k	15k
chlorpropham	0.0	0.5	0.9	18.4
Procymidone	0.5	0.0	0.7	8.6
Iprodione	0.2	0.4	3.1	4.9
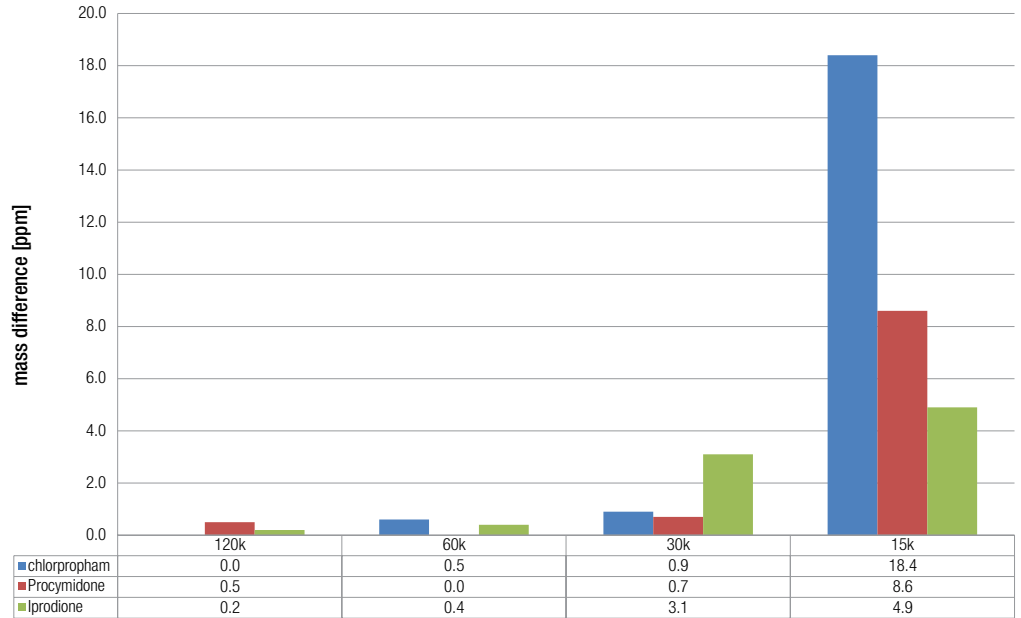
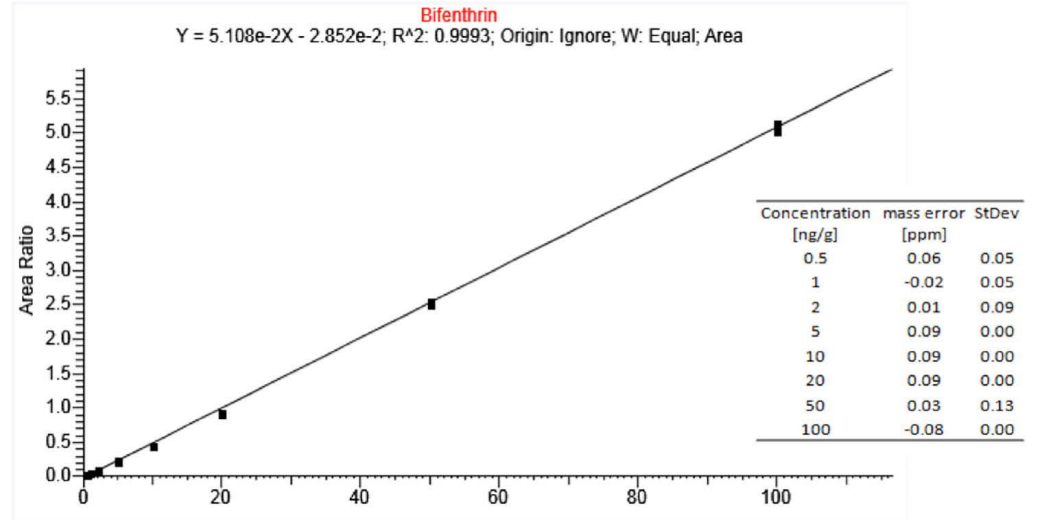## Consistent sub-ppm Mass Accuracy Across Peak Profile

The Q Exactive GC system provides routine stable mass accuracy of <1 ppm (internally calibrated) across the entire chromatographic peak, from the scans measured at the inflection points to the apex scans (Figure 4).

Figure 4. Scan-to-scan mass accuracy of 0.5 (RMS) of biphenyl ($C_{12}H_{10}$) identified in a carrot sample at 10 ng/g level. Data acquired at 60k (FWHM at *m/z* 200) resolution with >18 scans/peak (peak width 4 sec).
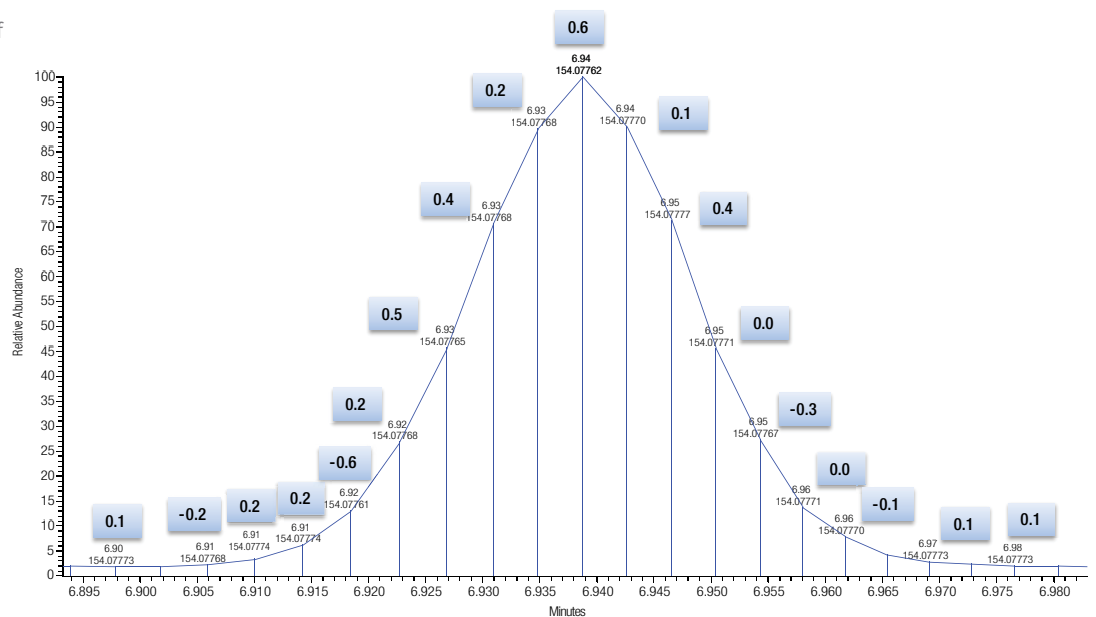
Moreover, accurate and precise mass measurements are consistently achieved at various target concentrations in complex matrix samples (Figure 5.)

Figure 5. Mass accuracy measurements for bifenthrin across a calibration curve (0.5–100 ng/g) prepared from matrix-matched baby food standards. The inset table shows the average values (n=3) of mass difference (± ppm) at each calibration level.

Consistent sub-ppm, mass-accurate measurements were observed when repeatedly injecting a baby food sample spiked with several pesticides at the 5 ng/g level (Figure 6). This result demonstrates the stability of system performance, which is critical when routinely analyzing complex samples, such as pesticide residues extracts.

Figure 6. Mass accuracy across n=14 repeat injections of a baby food sample spiked with several pesticides at 5 ng/g level.
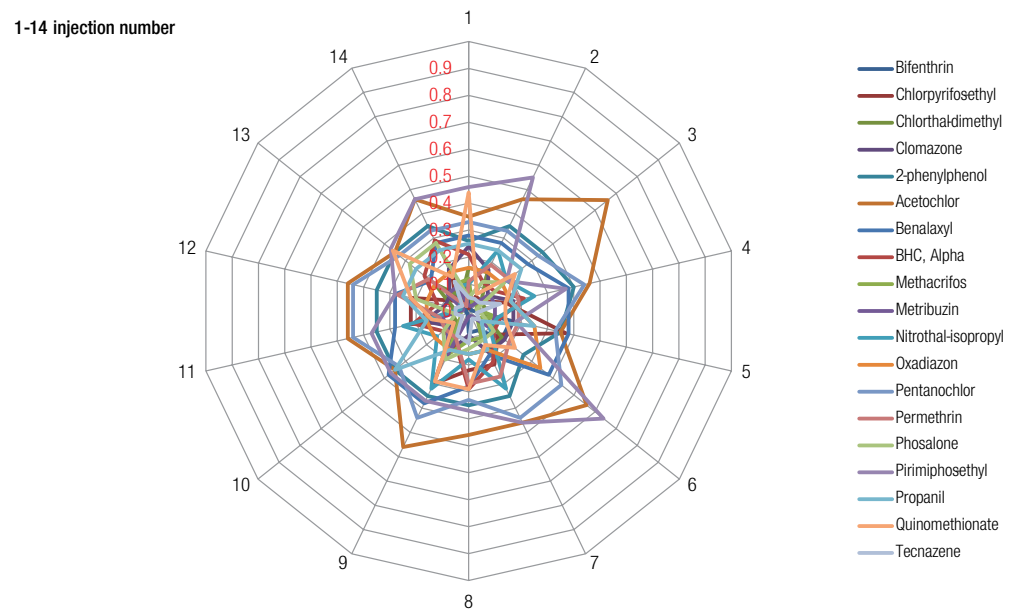
Linear and "In-spectrum" Dynamic Range

The linear and spectral dynamic range over which accurate measurements of a mass can be made is important for any accurate-mass analyzer. This range is particularly important for routine pesticide screening where the levels of the chemicals of interest are exceedingly high and/or very low and potentially co-elute with highly concentrated matrix components. The linear dynamic range of mass accuracy relates to the concentration range over which the ion of interest can be accurately measured. The "in-spectrum" dynamic range describes the ability of the instrument to measure an accurate mass for both low and high abundance ions within a mass spectrum; either related to the compound of interest or created by background components interfering. Until now, benchtop HR/AM GC-MS instrumentation has struggled to consistently provide a sufficiently high dynamic range in both cases. This failure may be related to operating with lower resolving power or to limitations in the detector used.

To enable optimal regulation of ions into the Orbitrap MS, a C-trap using automatic gain control (AGC), ensures that the mass measurement is accurate at all times. To test the concentration range over which the Q Exactive GC system can provide high mass accuracy measurements, an increasingly high concentration of hexachlorobenzene was injected into the system. The average mass accuracy for each ion over this range did not exceed 0.8 ppm mass error for each ion measured.

The range tested was greater than five orders of magnitude (0.1 ng/mL to 250,000 ng/mL). Table 2 provides the mass accuracy measurements taken for four of the ions produced by hexachlorbenzene.

Table 2. Mass accuracy greater than 5 orders of magnitude for four selected ions of hexachlorobenzene.

	Hexachlorobenzene ions			
	m/z 283.80962	*m/z* 248.84076	*m/z* 176.90601	m/z 141.93716
Concentration ng/mL	mass error (ppm)	mass error (ppm)	mass error (ppm)	mass error (ppm)
0.2	0.5	0.2	0.2	0.2
1	0.8	0.4	0.2	0.3
2.5	0.7	0.4	0.0	0.3
10	0.8	0.7	0.1	0.4
50	0.8	0.8	0.1	0.5
1000	1.0	1.0	0.3	0.7
5000	0.8	0.8	0.2	0.5
250000	0.8	0.5	0.0	0.4
Average mass error (ppm)	**0.8**	**0.6**	**0.1**	**0.4**

When evaluating the mass accuracy performance of the Q Exactive GC system in matrix, even at very low concentrations, when matrix components can potentially interfere with the target analytes, sub-ppm mass accuracy was obtained as exemplified below for dichlorvos measured in a fruit- and vegetable-based baby food sample (Figure 7). This data demonstrates the high in-spectrum dynamic range in terms of signal and mass accuracy.

Figure 7. Extracted full scan ion spectrum at RT = 8.5 min of a baby food sample spiked with dichlorvos at 0.5 ng/g. A close-up view of the measured ion cluster for m/z 184.97650 corresponding to $C_4ClH_7O_4P$ versus the theoretical isotopic cluster showing excellent mass accuracy and isotopic pattern match.
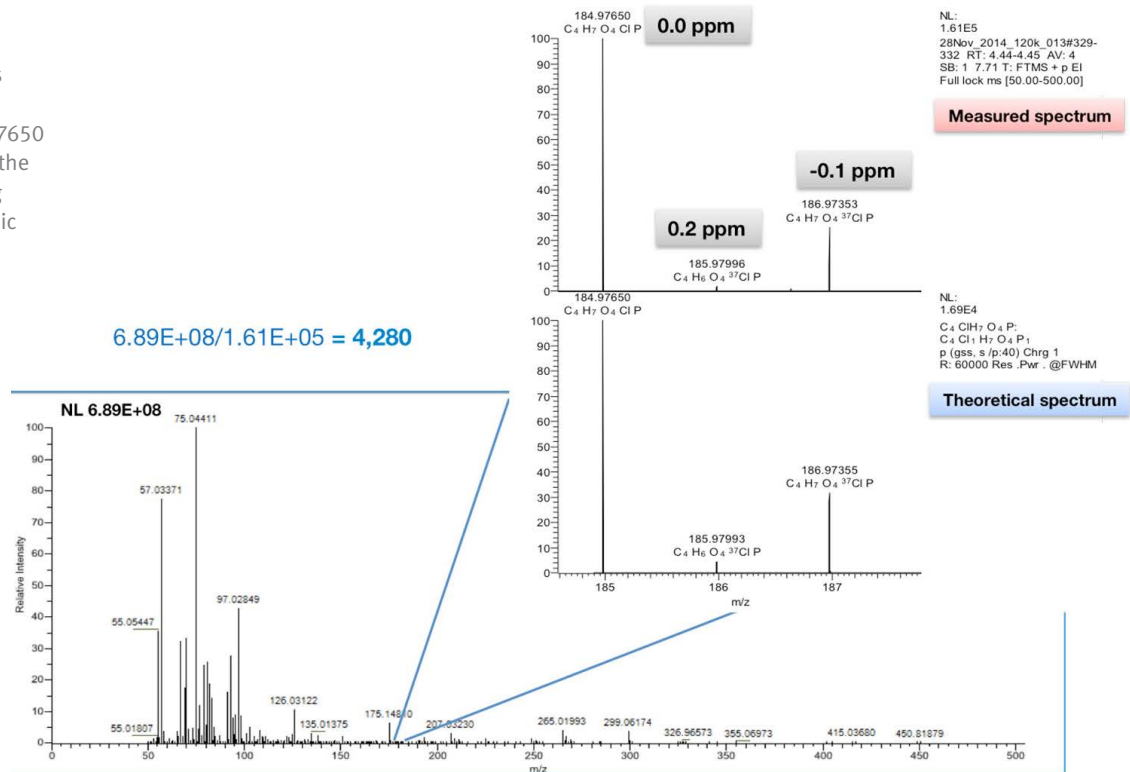
Mass Accuracy Across Mass Range

Sub-ppm mass accuracy measurements were obtained across a mass range of 69–960 Da, as demonstrated in Figures 8 and 9.

Figure 8. Mass accuracy across mass range (69–960 Da) measured from calibration gas ions (FC43) and the molecular ions of tri-BDE through deca-BDE from single injection.
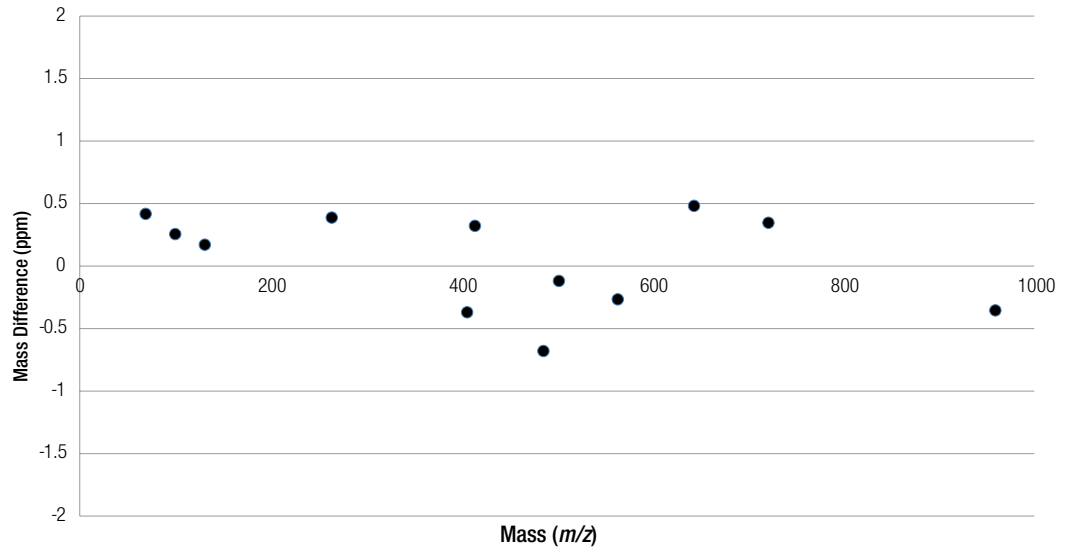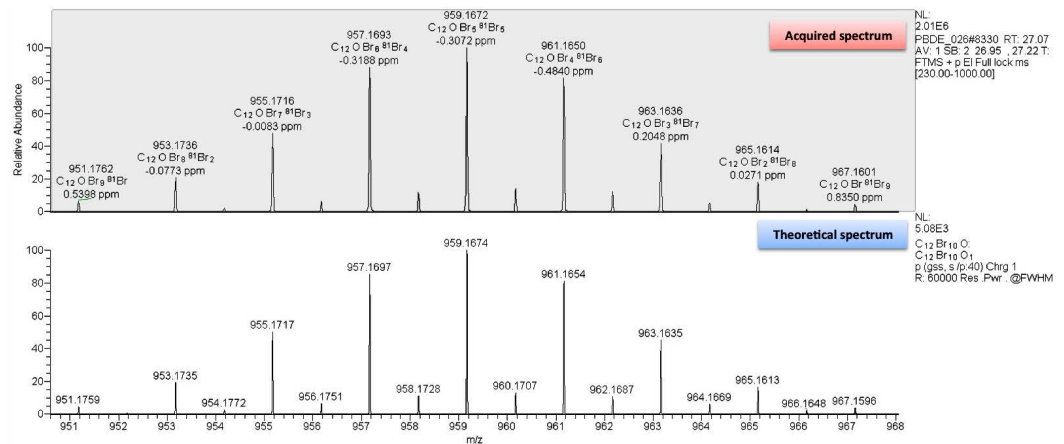

Figure 9. Mass accuracy (0.6 ppm RMS) of high masses shown for deca-BDE molecular ion cluster ($C_{12}Br_{10}O$). Data acquired in full scan at 60k resolving power.

Conclusions

The data presented in this work demonstrate that the Q Exactive GC mass spectrometer delivers routine accurate mass measurements over a range of analytical conditions.

Consistent mass accuracy is achieved across a chromatographic peak, at low and high concentrations, at low and high masses, and for various compound classes and matrix types.

The high resolving power of the system facilitates outstanding mass accuracy, and this combination can be used in a number of application areas where high confidence in compound discovery, screening, identification, and quantification is required.

Acknowledgements

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