GC/MS Fundamentals:
What are GC/SQ, TQ, and Q-TOF and When to Use Them?

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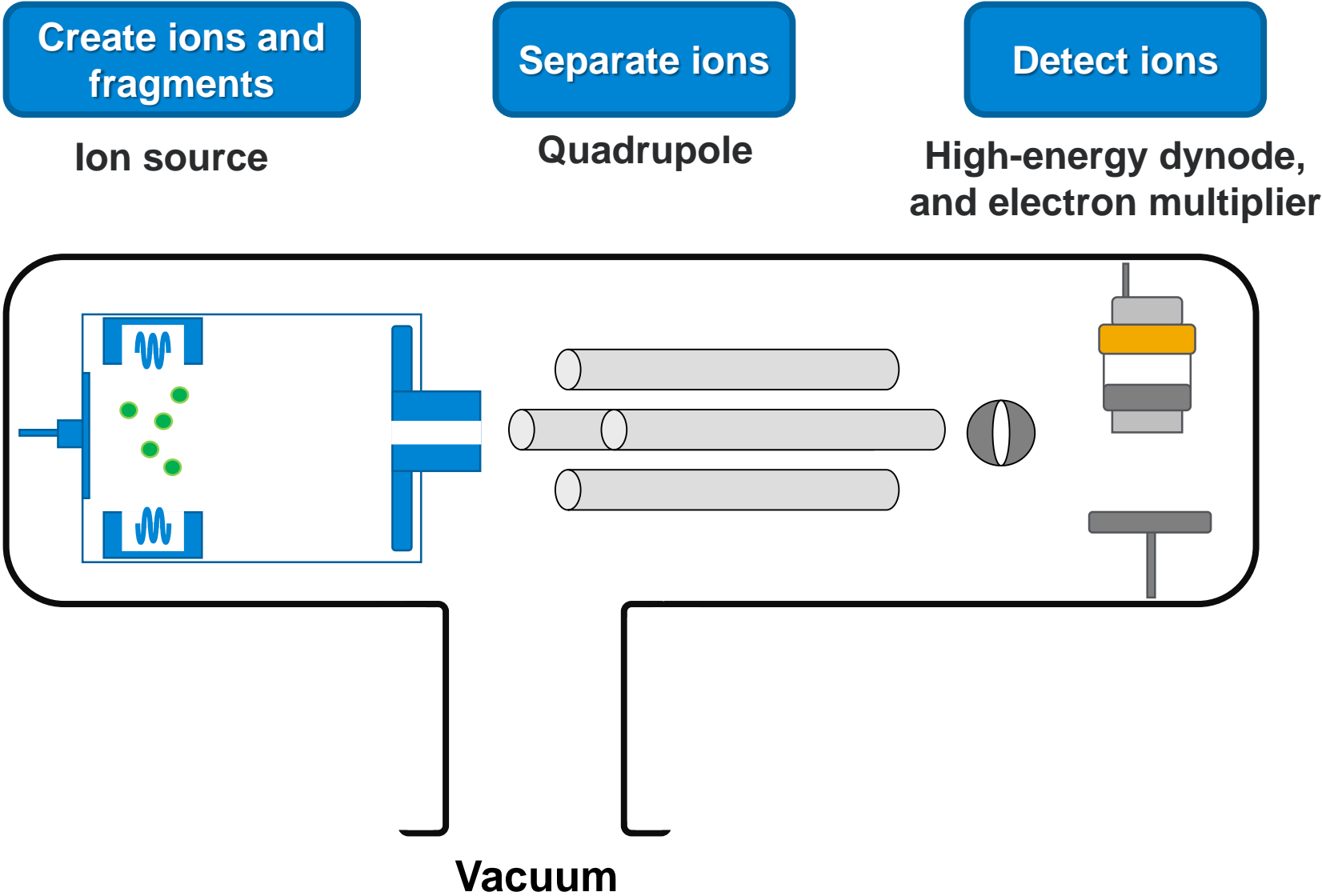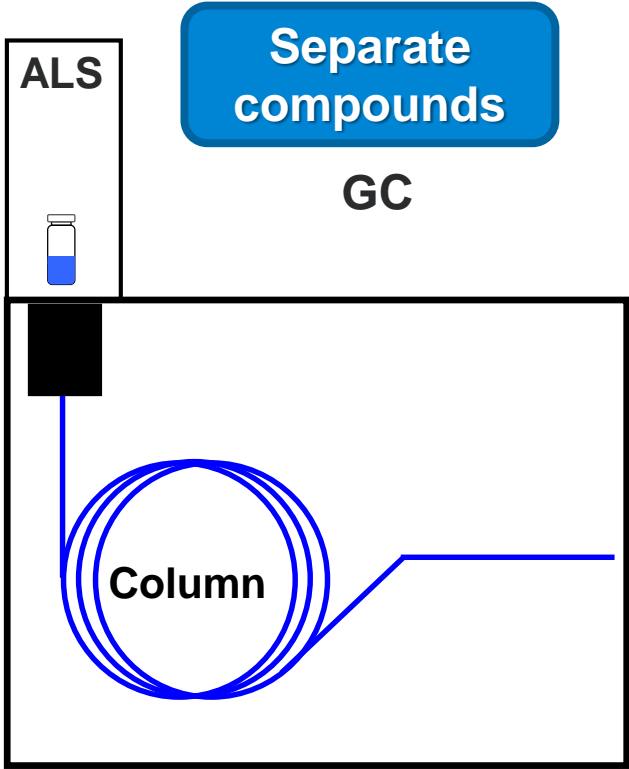
Objectives

Overview of how Agilent's GC/MS systems work:
- GC/single quad (GC/MSD SQ)
- GC/triple quad (GC/MS TQ)
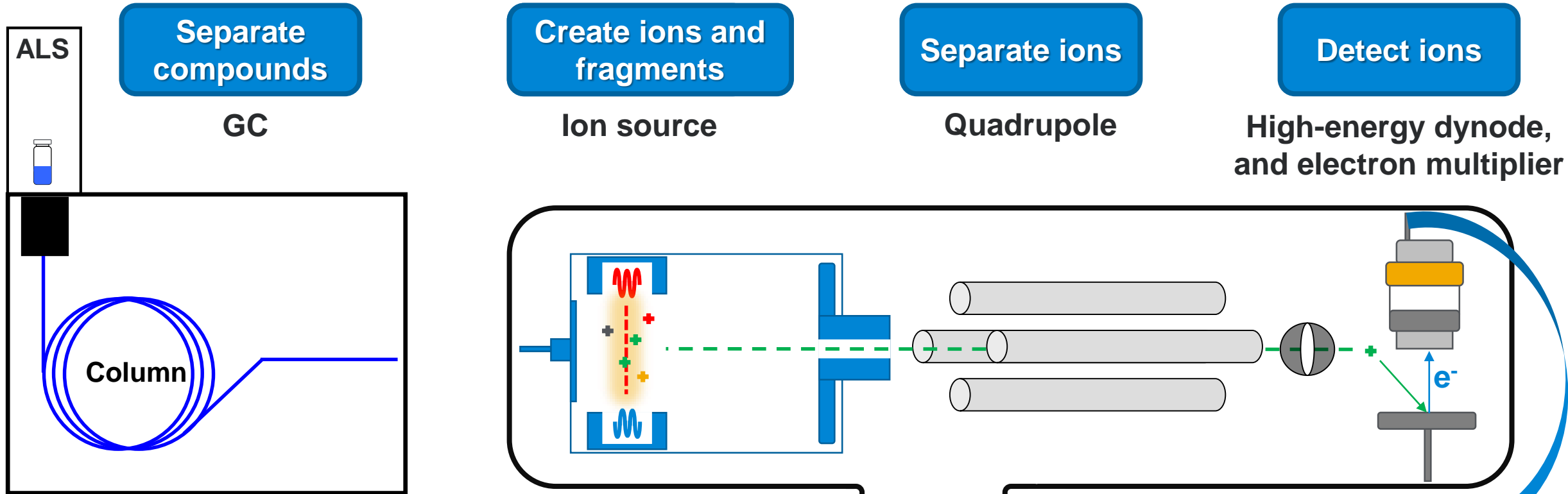- GC/quadrupole time-of-flight (GC/MS Q-TOF)

Overview of how customers use each type to solve their analytical problems
- Example: Pesticides in essential oils

SQ GC/MSD

Separate compounds

GC

Create ions and fragments

Ion source

Separate ions

Quadrupole

Detect ions

High-energy dynode, and electron multiplier

ALS

Column

Vacuum

SQ GC/MSD

Separate compounds

GC

Create ions and fragments

Ion source

Separate ions

Quadrupole

Detect ions

High-energy dynode, and electron multiplier

ALS

Column

e⁻

Vacuum

During the run, quadrupole is (for example) scanned from 35 m/z to 500 m/z at 3.2 scans/sec. Signal that reaches the detector is recorded in the data file.
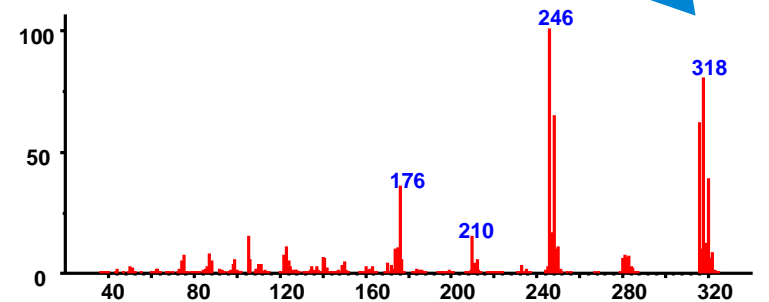
SQ GC/MSD: Example Spectrum at 24.065 Minutes

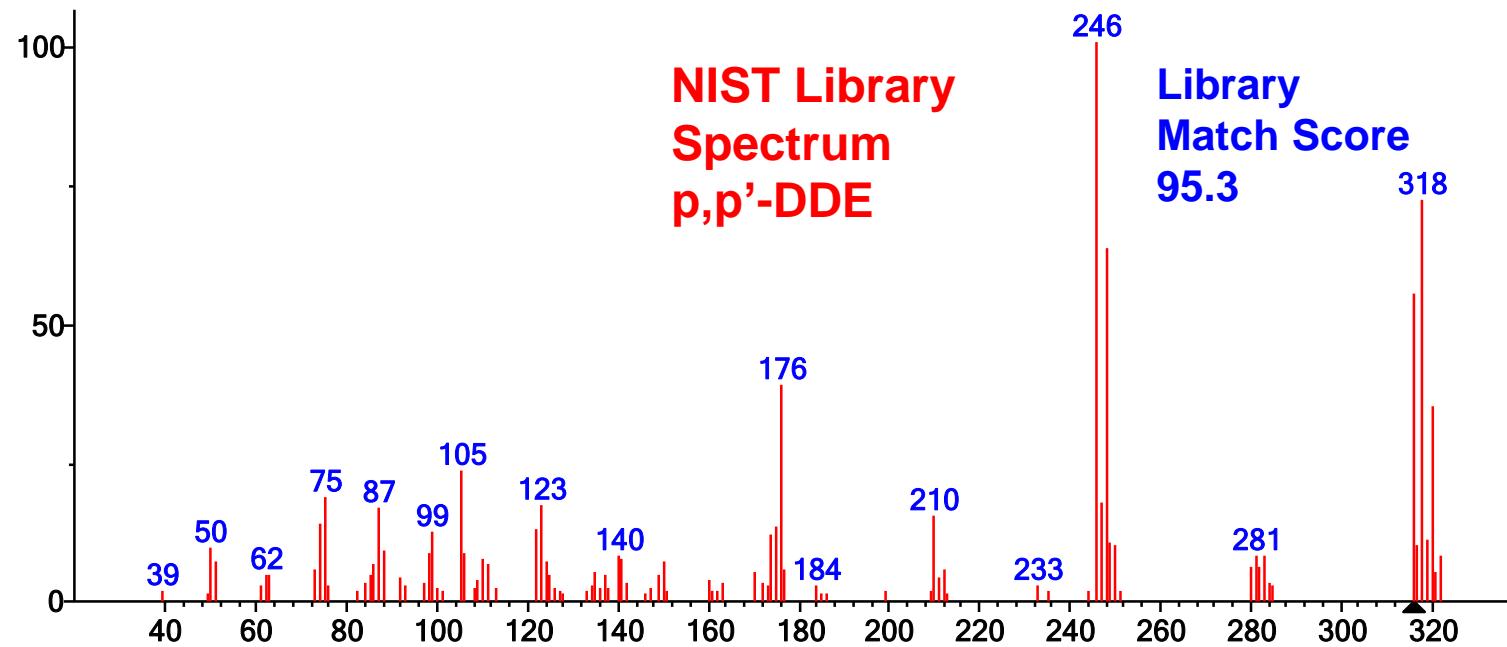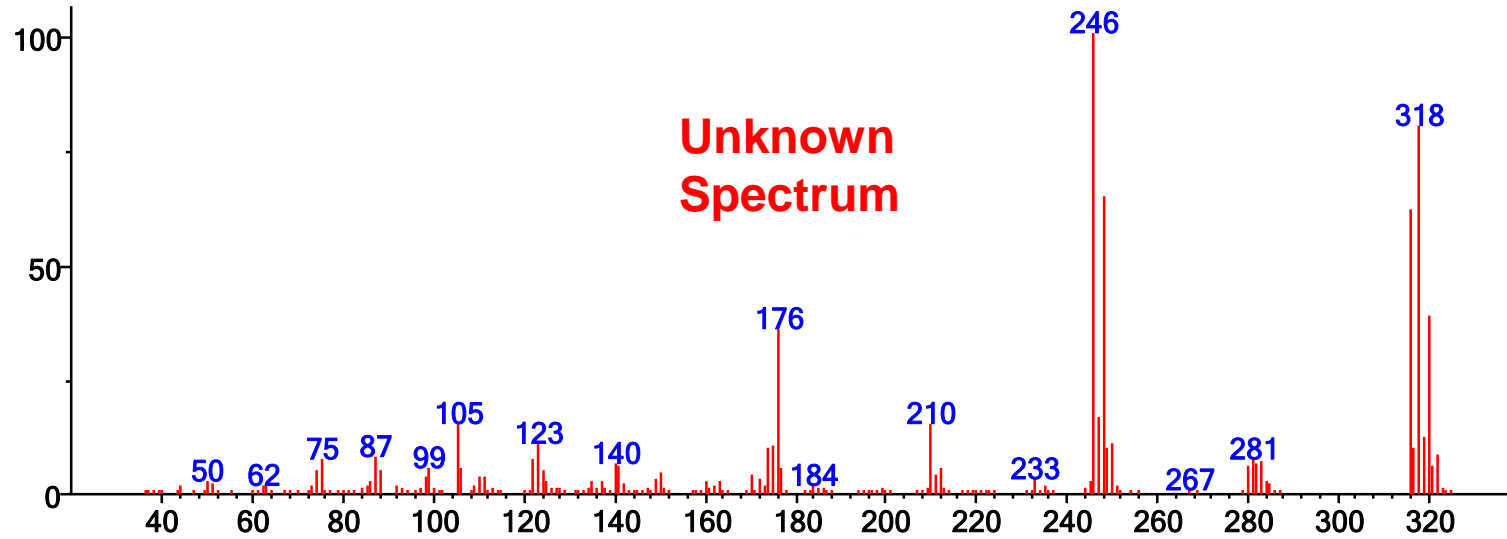Agilent Technologies   For Research Use Only

Agilent
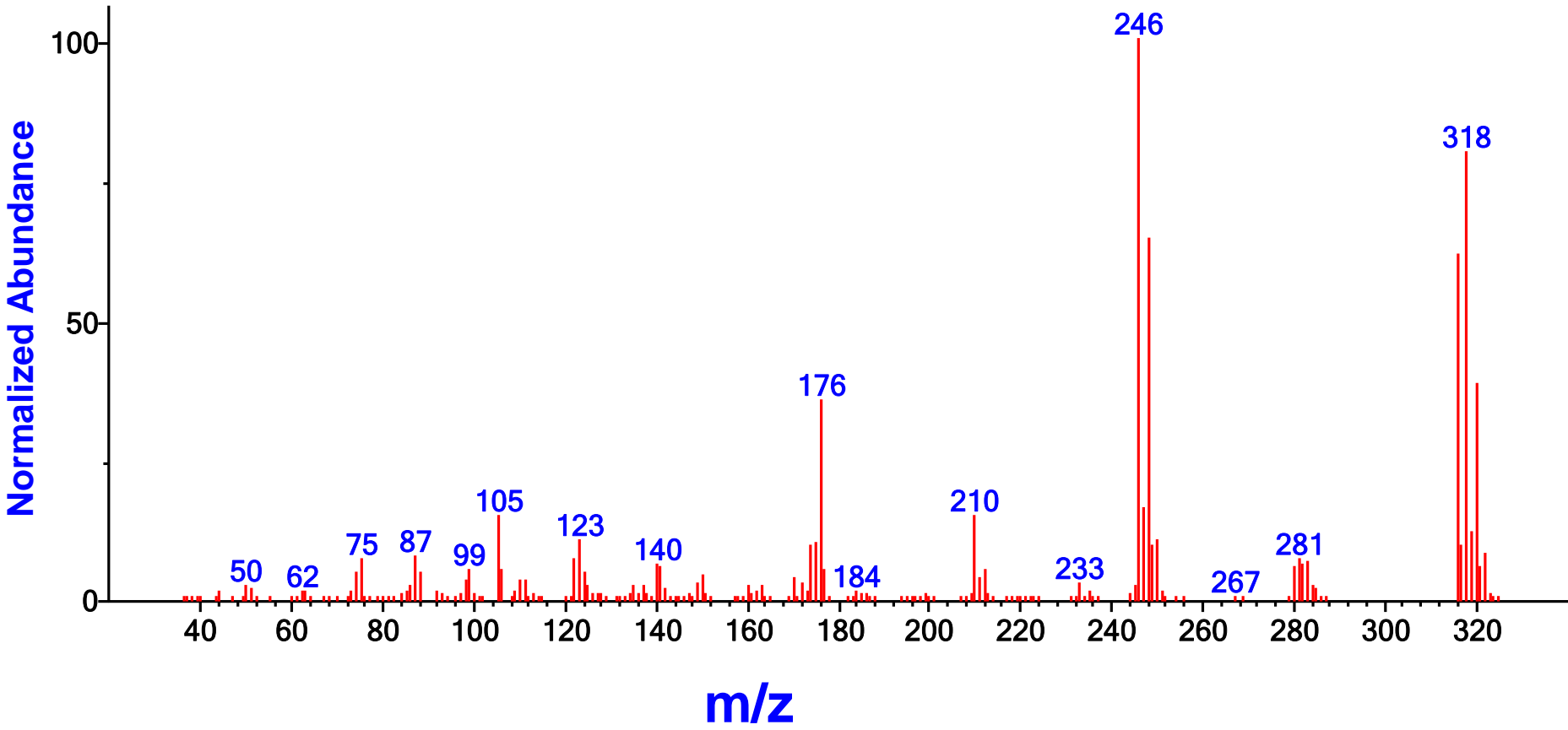
SQ GC/MSD: Processing SQ Scan Data

Scan data is processed to create:

- Total Ion Chromatogram (TIC)
- Base Peak Chromatogram (BPC)
- Extracted Ion Chromatograms (EIC)
- Spectra for library searching
- Spectra for unknown identification
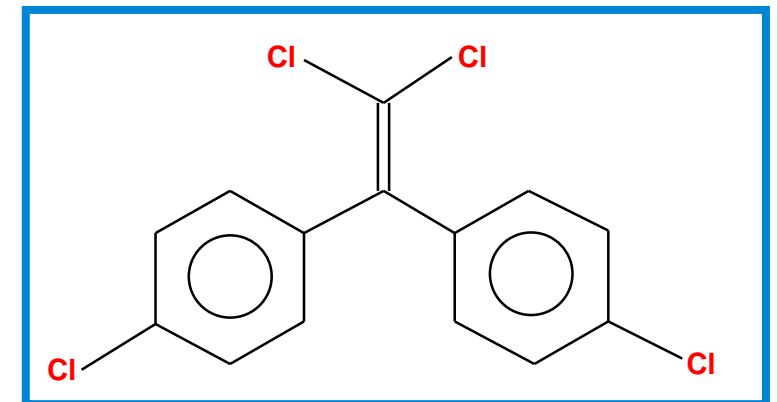
Agilent Technologies   For Research Use Only
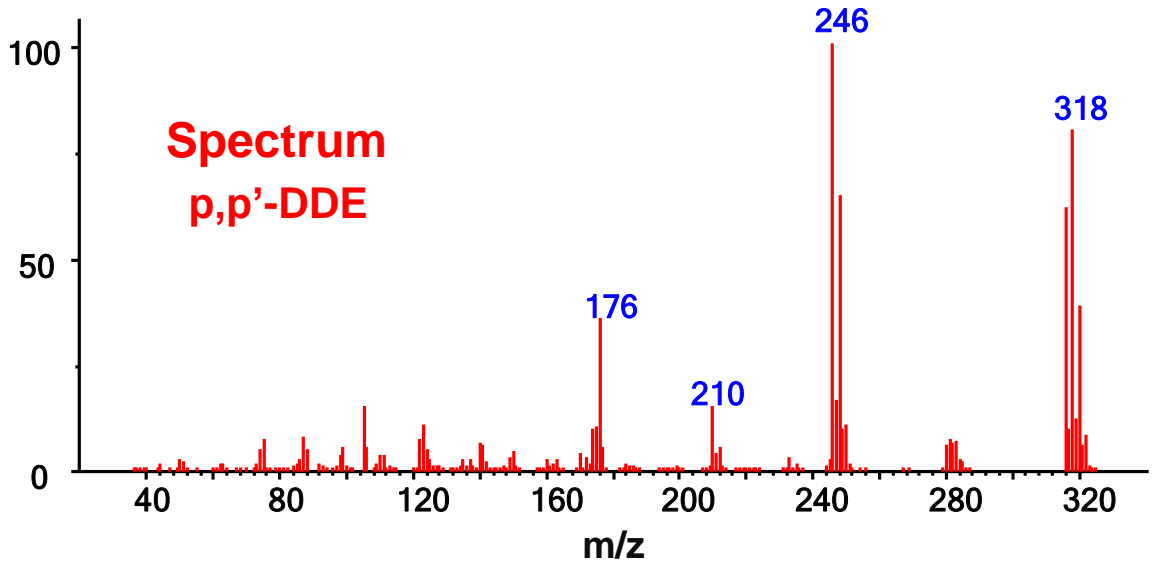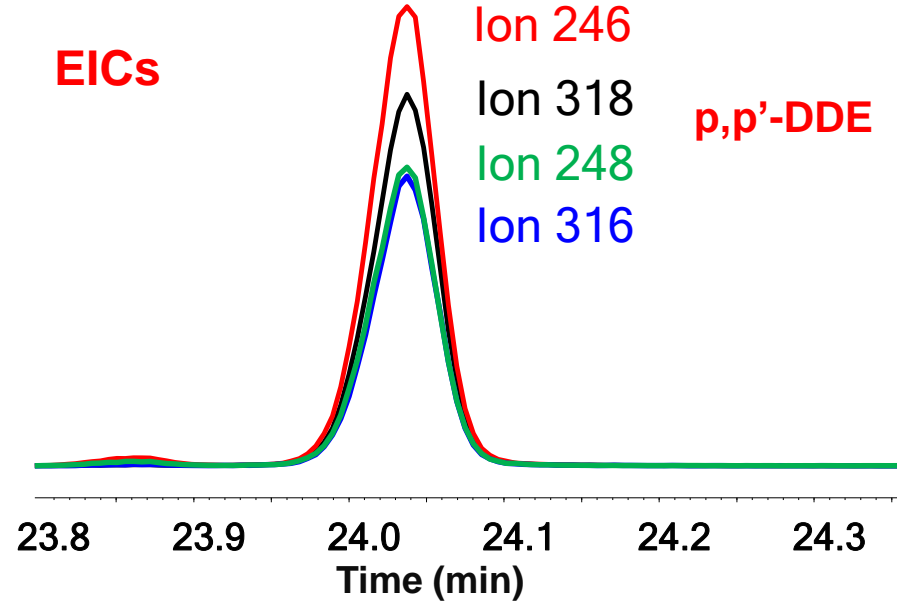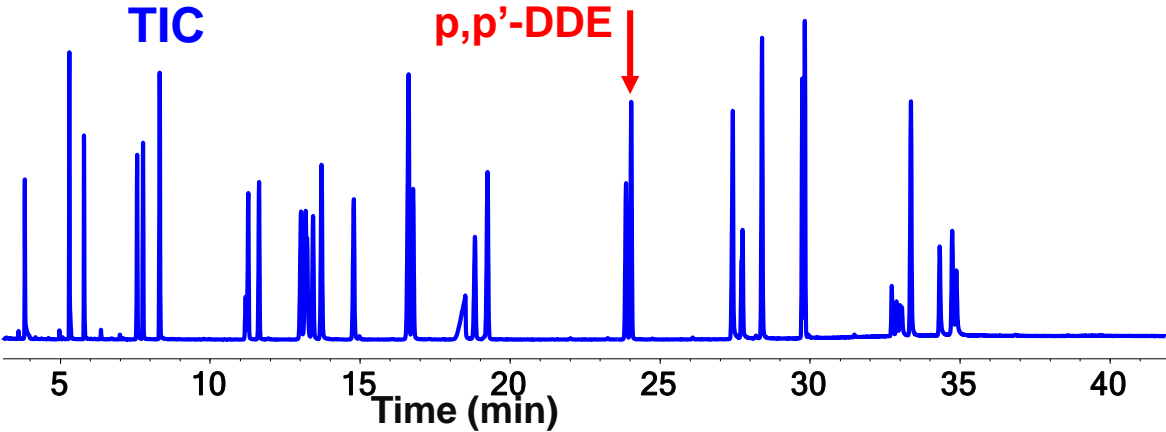
# SQ GC/MSD: Library Searching SQ Scan Data

Searching unknown spectra against NIST or other libraries is widely used for identification of compounds.

Spectral deconvolution software from NIST (AMDIS) and now MassHunter SureTarget can be used to produce spectra with the ions from overlapping matrix peaks removed.

SIM: Single Ion Monitoring

Full scan spectrum 35–500 amu

SIM monitoring 8 Ions instead of 465
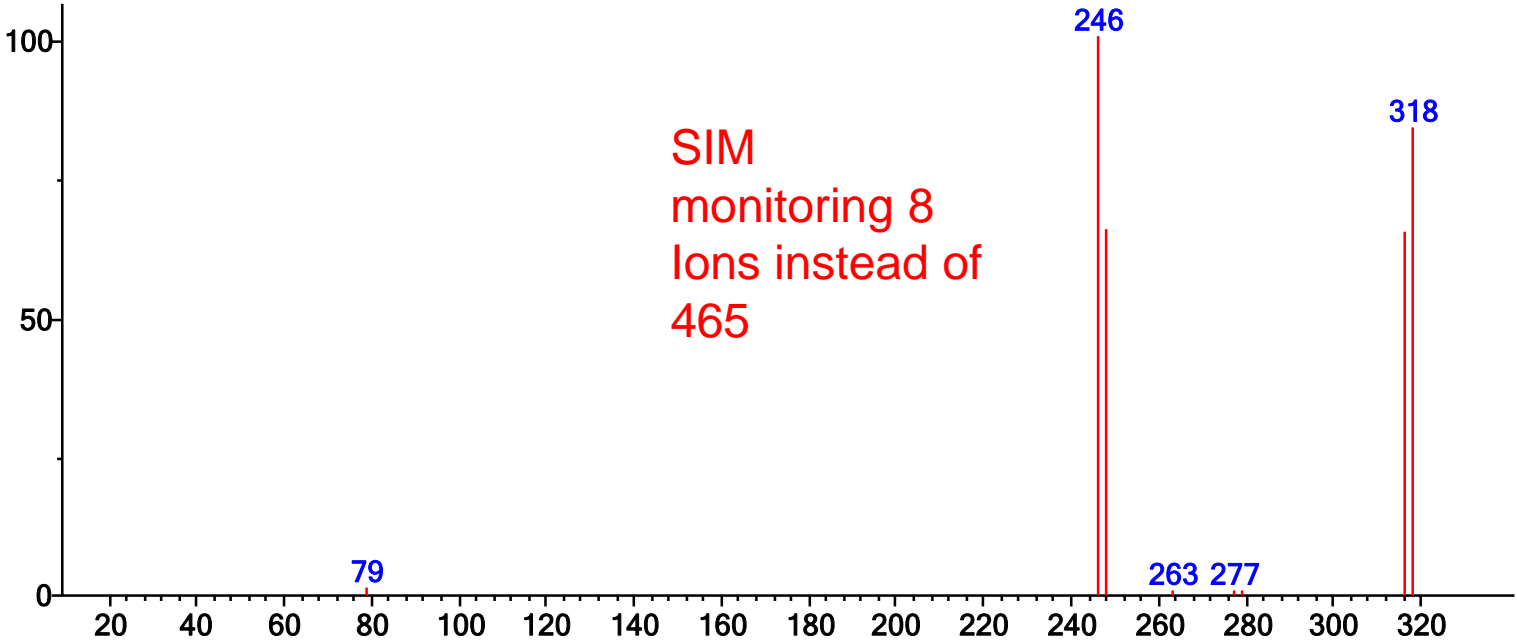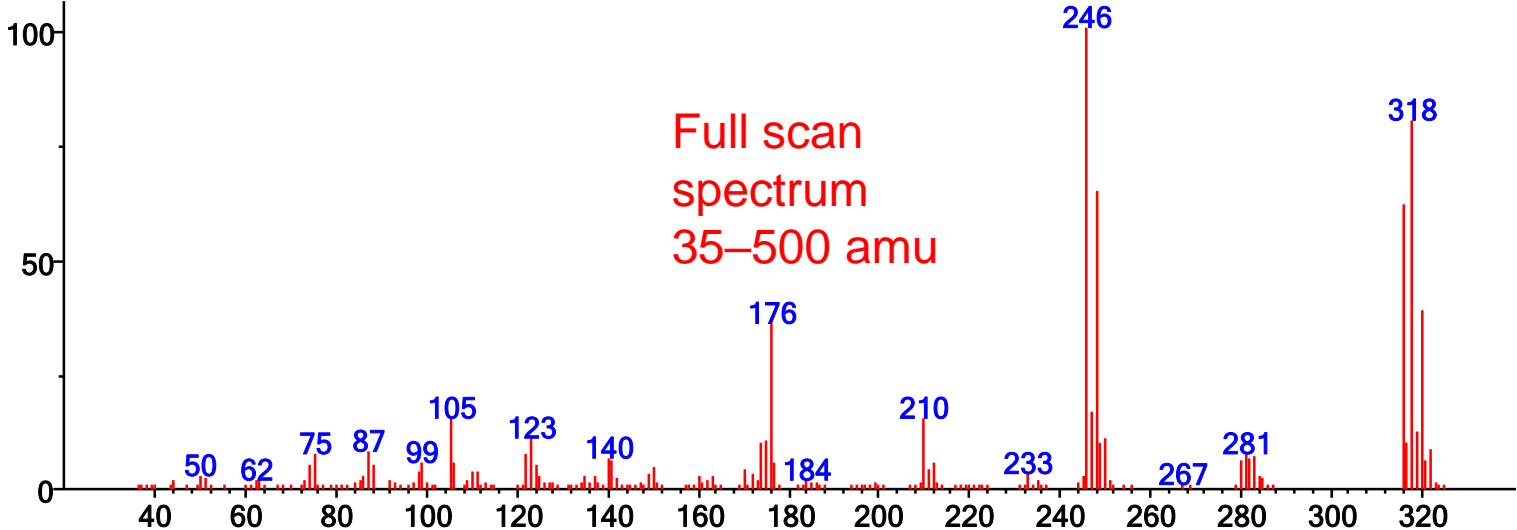
During chromatographic run, quad is programmed to monitor a small number of ions for specific analytes in specific time ranges.

More time is spent measuring each ion vs scan.

This gives ~10 x better signal-to-noise and better area precision. However, ability to search spectra for unexpected unknowns is lost.

SQ GC/MSD: Limited Selectivity of SQ

TIC Spinach Extract

EIC 246 amu

p,p'-DDE

matrix interferences at m/z 246
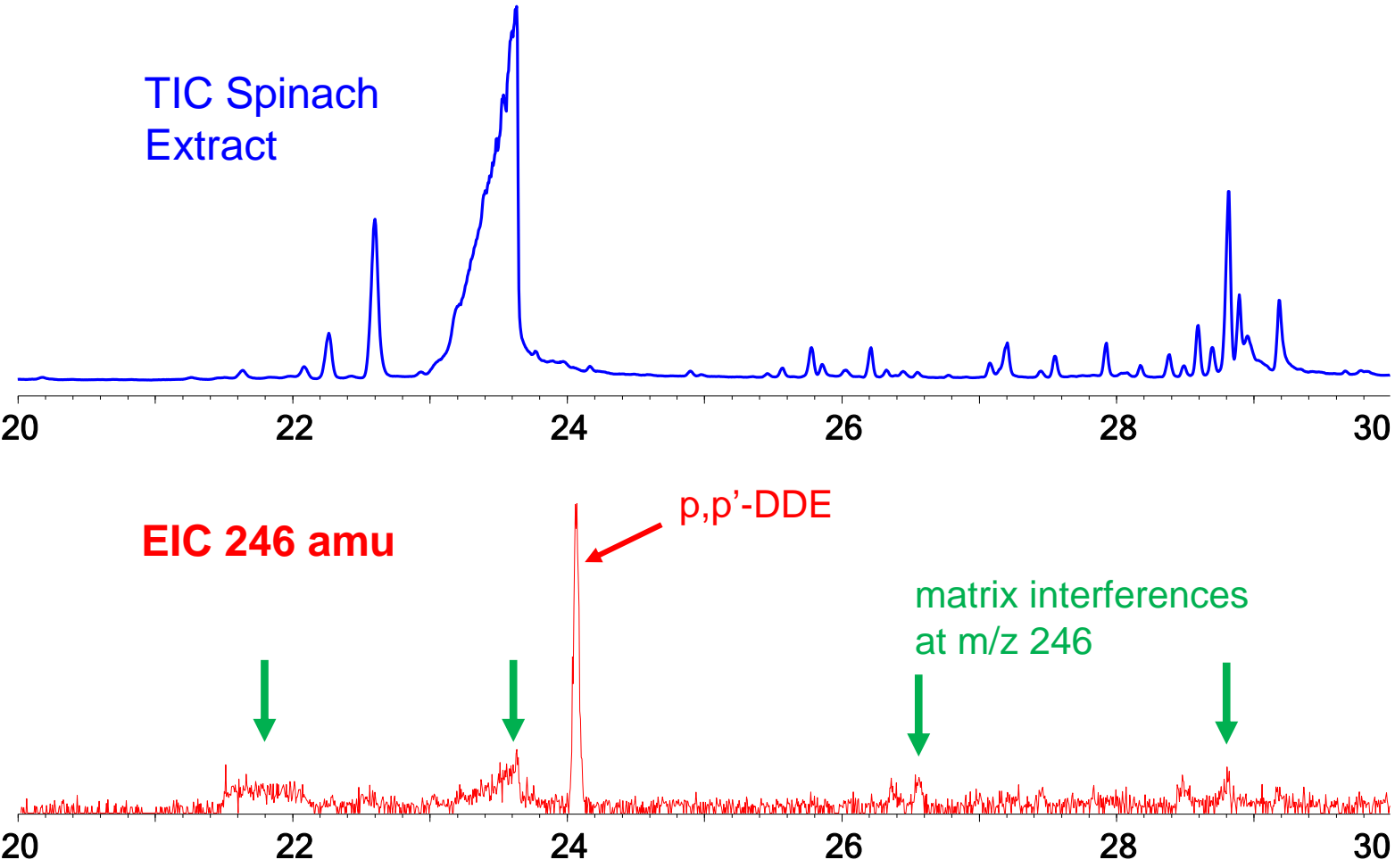
The p,p'-DDE peak is visible in this spinach extract, but the baseline shows that the SQ has only limited selectivity over other matrix compounds that also have m/z 246 as a fragment.

This limits the ability of the SQ to find traces of pesticides in extracts with high levels of matrix interferences.

SQ GC/MSD: Summary

Most widely used GC/MS technique
- Least complicated
- Relatively rugged
- Alternative to traditional GC detectors in many applications

Modes of use
- Quantitative analysis (how much is in there?) using target ion and 1–3 qualifiers
  - SIM: best S/N (detection limit), but limited number of analytes and no spectra for identifications
  - Scan: ~10 x poorer S/N, unlimited number of analytes and spectra for identifications
- Qualitative analysis (what is in there?)
  - Scan: Searching of deconvoluted spectra against libraries like NIST, Agilent RTL libraries, or others
  - Retention time versus response chromatogram like any GC detector

Limitations
- Selectivity over interferences from matrix
- Selectivity over some GC detectors (uECD, FPD, NPD, SCD, etc.)
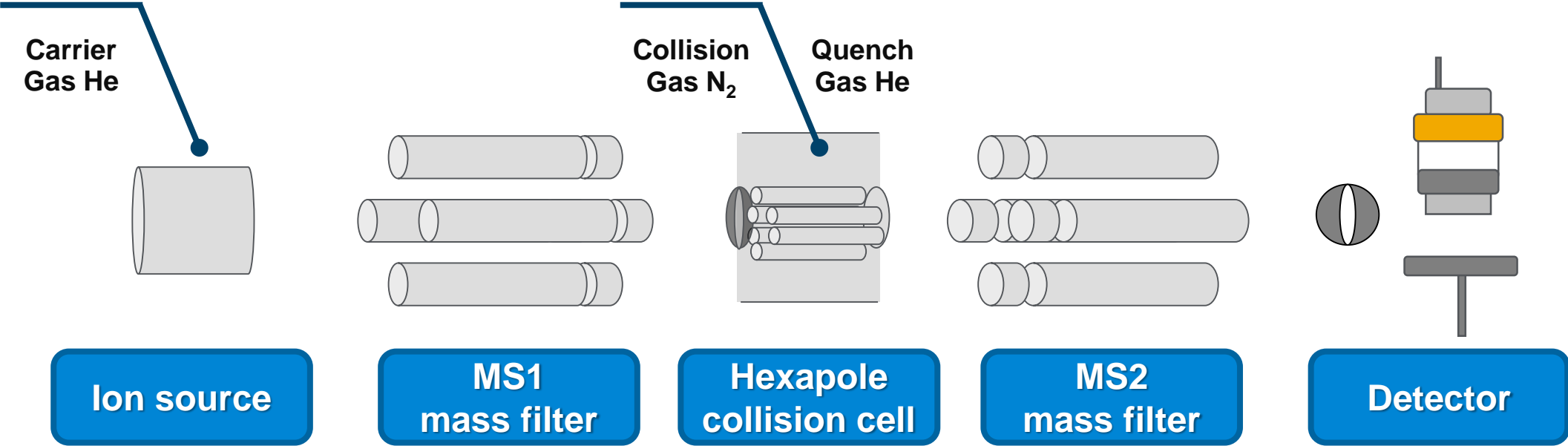- Unit mass resolution limits spectral interpretation of unknowns

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Agilent GC TQ Systems

**7000D
extractor source**

**7010B
HES source**

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TQ GC/MS: What is MS/MS Analysis?

MS/MS requires multiple mass filters in series

Carrier Gas He	Collision Gas N_2	Quench Gas He

Ion source	MS1 mass filter	Hexapole collision cell	MS2 mass filter	Detector

Typical analysis	=	Selection of precursor ion (MS1)	Fragmentation of precursor ion (Collision cell)	Selection of fragment ions (MS2)	Detection

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TQ GC/MS: Multiple Reaction Monitoring (MRM)

Step 1: Precursor ion of target passed by MS1. Contains fragment ions of mass 210 created from **both** analyte **and** interference molecules.
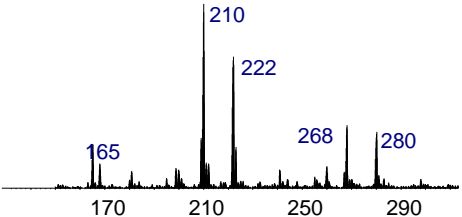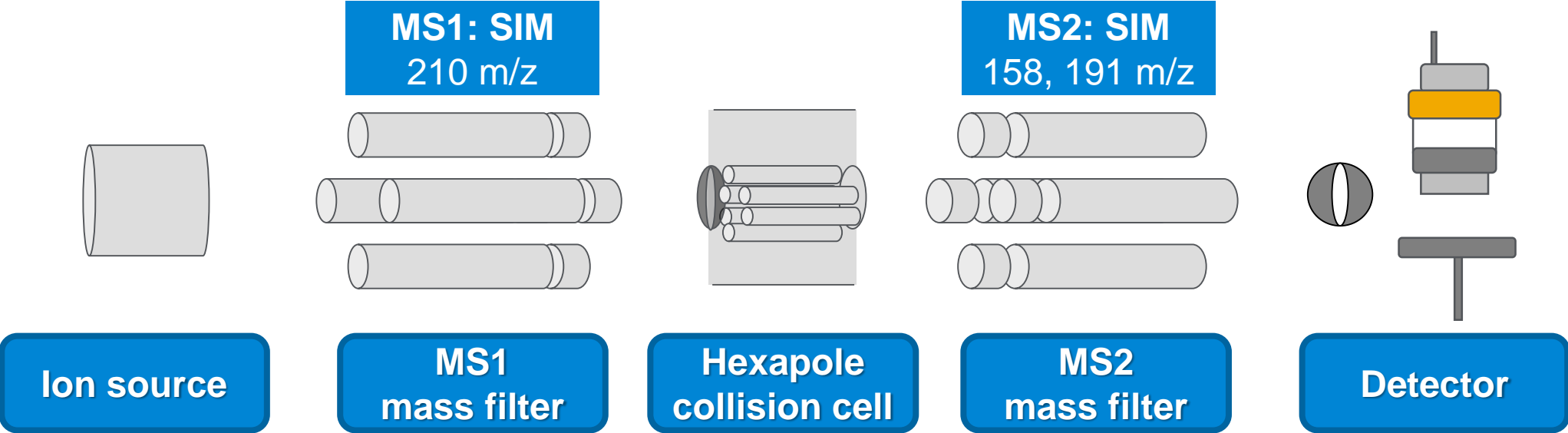
MS1: SIM 210 m/z		MS2: SIM 158, 191 m/z	

Ion source	MS1 mass filter	Hexapole collision cell	MS2 mass filter	Detector

Spectrum with background ions
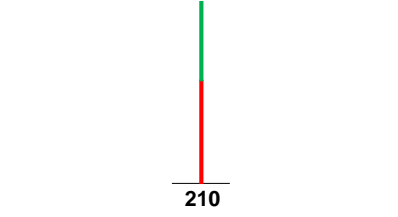
210
222
165
268
280
170   210   250   290

210
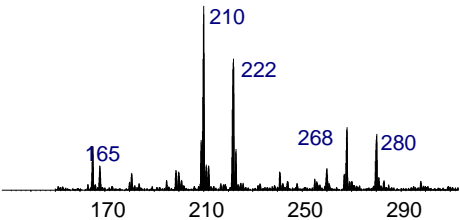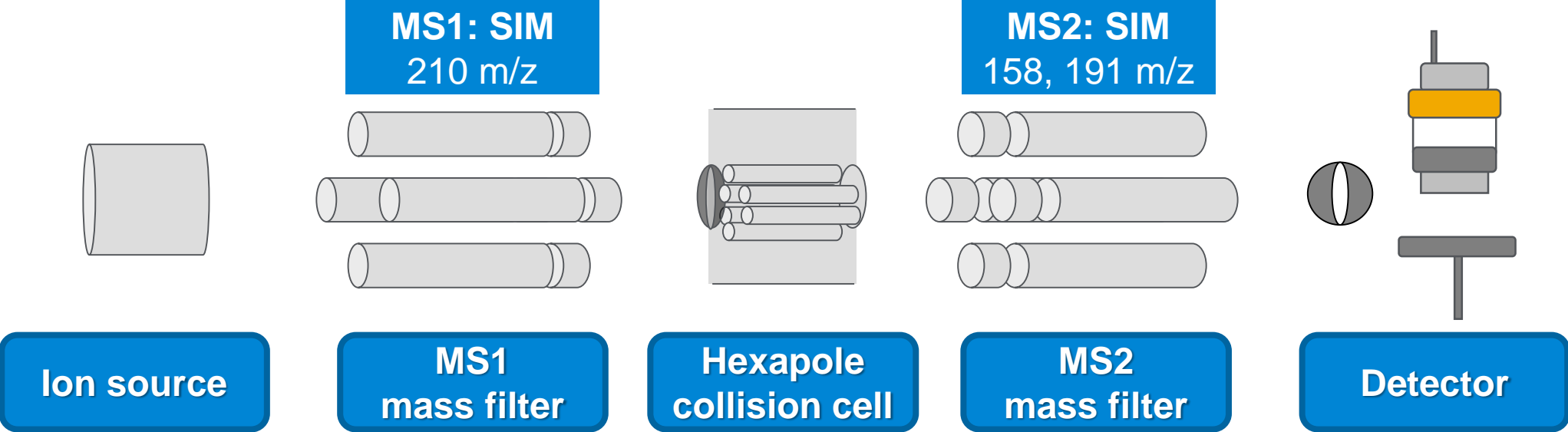
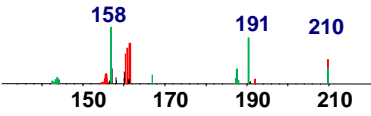MS1 lets only target ion 210 pass through

TQ GC/MS: Multiple Reaction Monitoring (MRM)

Step 2: The collision cell generates ions from the mass 210 fragments from **both** analyte **and** interferences.

| MS1: SIM 210 m/z | | MS2: SIM 158, 191 m/z | |

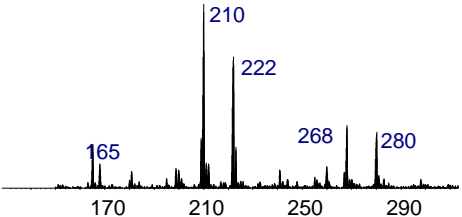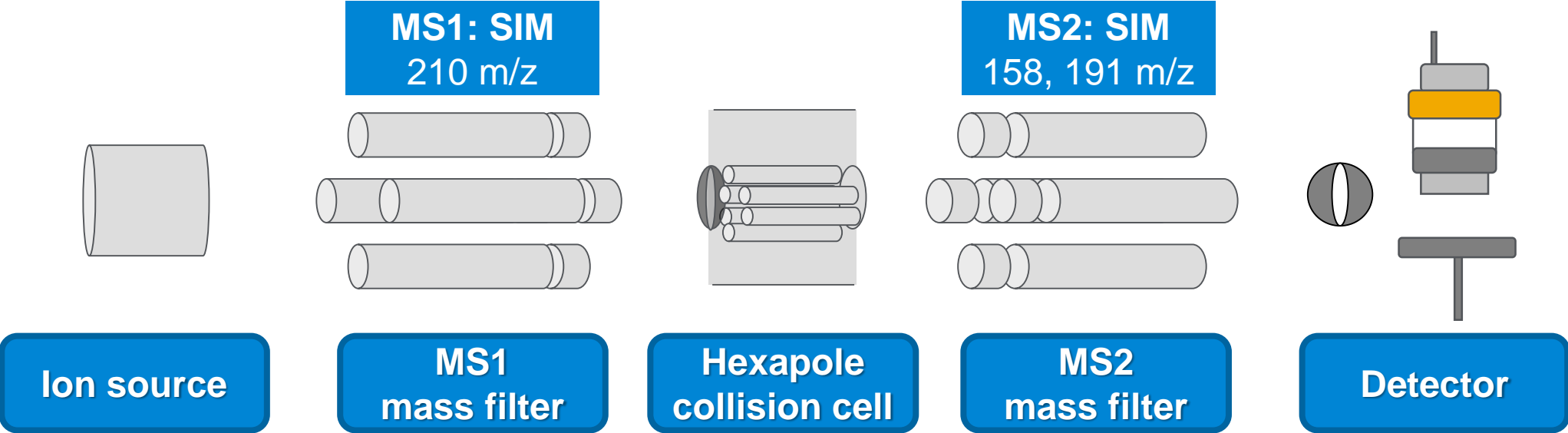| Ion source | MS1 mass filter | Hexapole collision cell | MS2 mass filter | Detector |

Spectrum with background ions
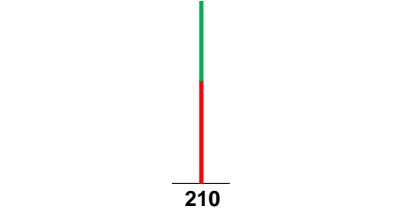
MS1 lets only target ion 210 pass through

Collision cell breaks ion 210 into fragments

TQ GC/MS: Multiple Reaction Monitoring (MRM)

Step 3: MS2 is set to filter for quantification and qualifier product ions **only** formed by the analyte (unique ions.)

MS1: SIM 210 m/z

MS2: SIM 158, 191 m/z

Ion source

MS1 mass filter

Hexapole collision cell

MS2 mass filter

Detector

Spectrum with background ions

MS1 lets only target ion 210 pass through
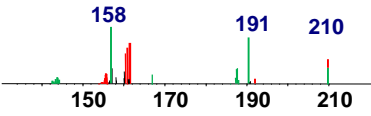
Collision cell breaks ion 210 into fragments
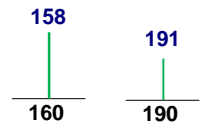
MS2 monitors only characteristic fragments 158 and 191 created from ion 210, for quant and qual.

Comparison of SQ SIM and TQ MRM data: p,p'-DDE, 10 ppb, various matrices

SIM (246)   MRM (246→175)

Apple   S/N = 448

Cabbage   S/N = 241

Ginseng   S/N = 446

Orange   S/N = 456

Spinach   S/N = 260

(All injections = 1 µL)

TQ GC/MS: Summary
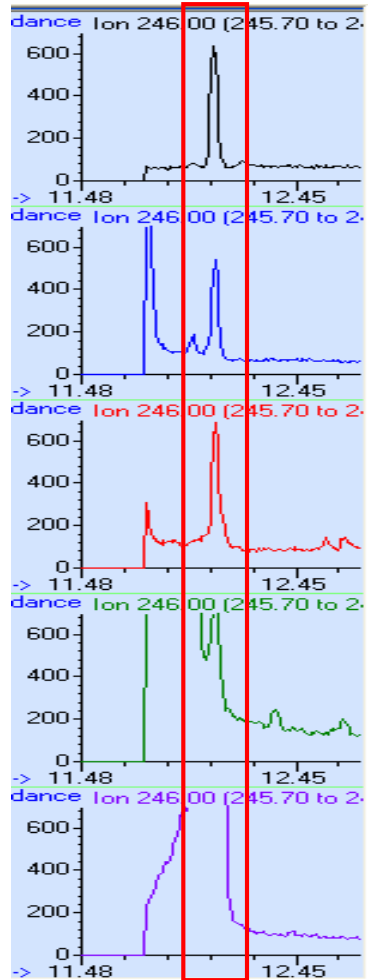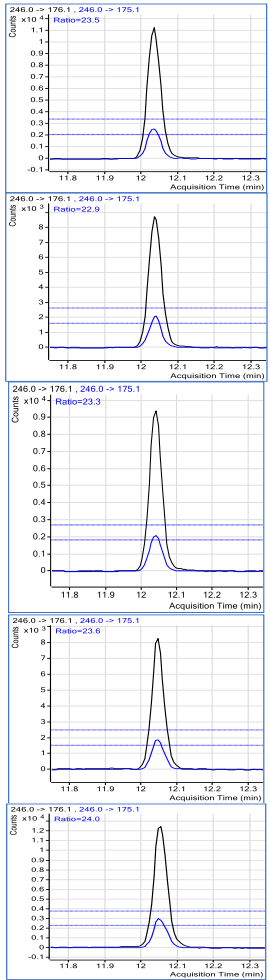
Most widely used GC/MS technique for target analysis of pesticides
- More complicated method development choosing precursor and product ions and collision voltages
- MRMs provide extremely high selectivity necessary in complex matrices

Modes of use
- Quantitative analysis (how much is in there?)
  - MS2 SIM similar to SQ SIM
  - MRM using target ion and 1-2 qualifiers
- Qualitative analysis (what is in there?)
  - Scan: Searching of deconvoluted spectra against libraries like NIST, Agilent RTL libraries, or others

Limitations
- Unit mass resolution limits spectral interpretation of true unknowns
- Limited number of MRMs per run
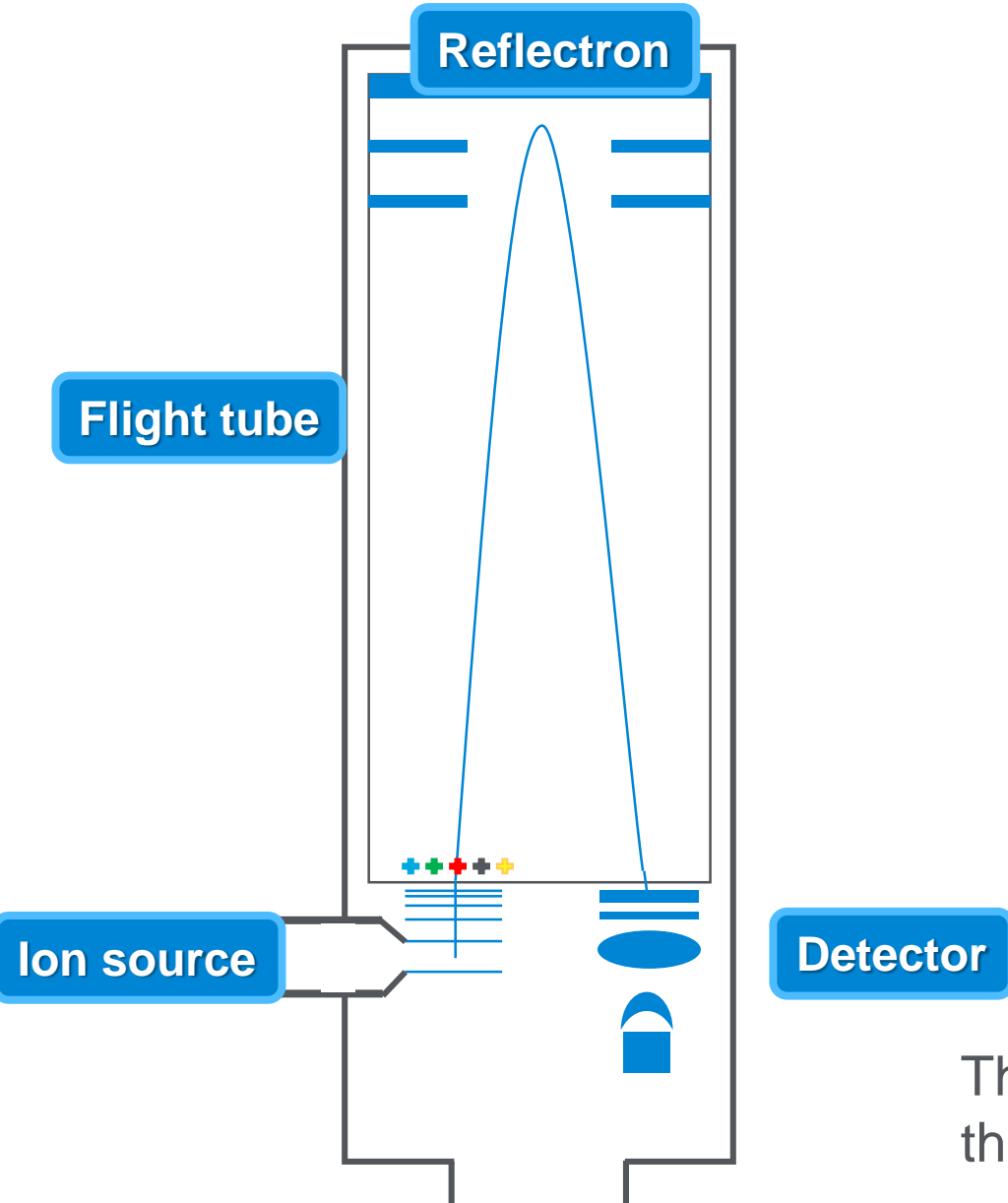- Scan mode useful, but not quite as good as SQ in S/N or spectral match to NIST

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Q-TOF GC/MS

7200 Q-TOF
Extractor Source

7250 Q-TOF
HES Source

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Q-TOF GC/MS: What is Time of Flight?

Flight tube

Ion source

Detector

$$K_e = \frac{1}{2}\, mv^2$$

The kinetic energy, K_e, of ions as they are pulsed in the flight tube, where

m = mass
v = velocity

$$v = L/t$$

The ion velocity equals the flight path (L) divided by (t) time
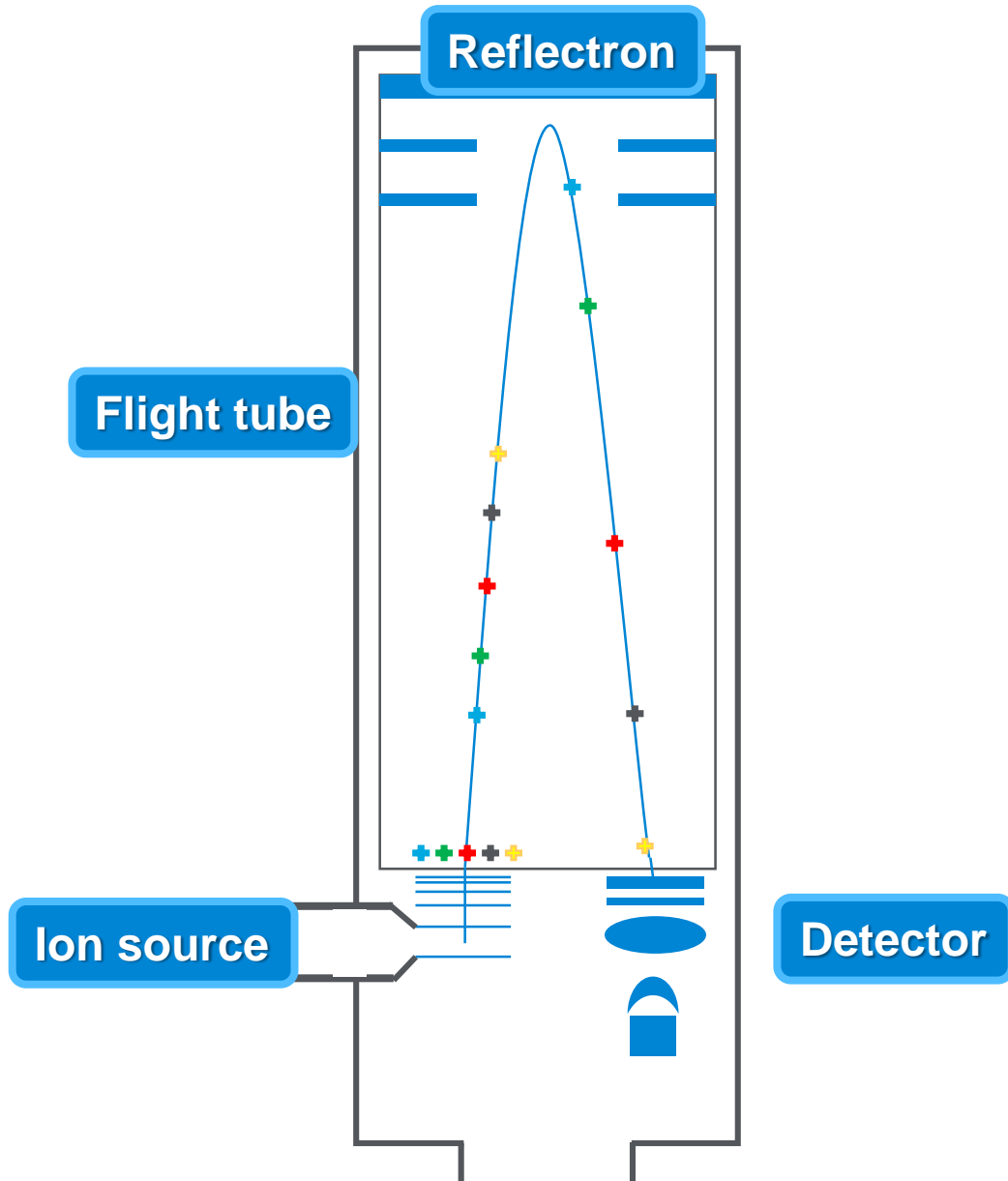
$$\frac{L}{t} = 1/2mv^2$$

Combine equations…

$$m = t^2(2Ke)/L^2$$

$(2Ke)/L^2$ is considered constant

$$m \propto t^2$$

The higher the m/z for an ion, the longer it takes to transit the flight tube and reach the detector

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Q-TOF GC/MS: Transients


1. Pulse ions every 100 microseconds.

2. Measure at detector each nanosecond.

3. 100,000 data points in each transient.

4. Sum 2000 – 10,000 transients into one spectrum.

Produces spectra with excellent ion statistics.

Transient—one packet of ions pulsed down the flight tube and detected. Data from many pulses, typically ~10,000 transients, are summed to create a spectrum.

Q-TOF GC/MS: MS or TOF mode
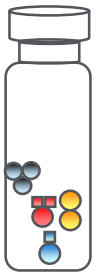
GC
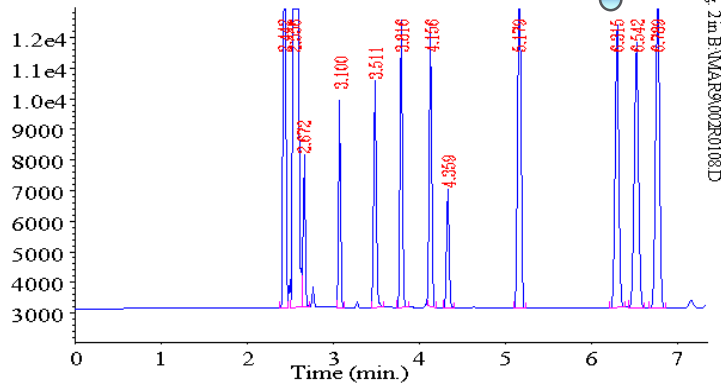
TOF mode can provide the accurate mass of the molecular ion, when available.

Similar to EI scan mode spectra from a SQ.

④ **TOF – Separation of ions by time**

② **Total ion transmission through quadrupole**

⑤ **Detection and digitalization**

① **Ionization in source**

③ **No energy applied in collision cell**

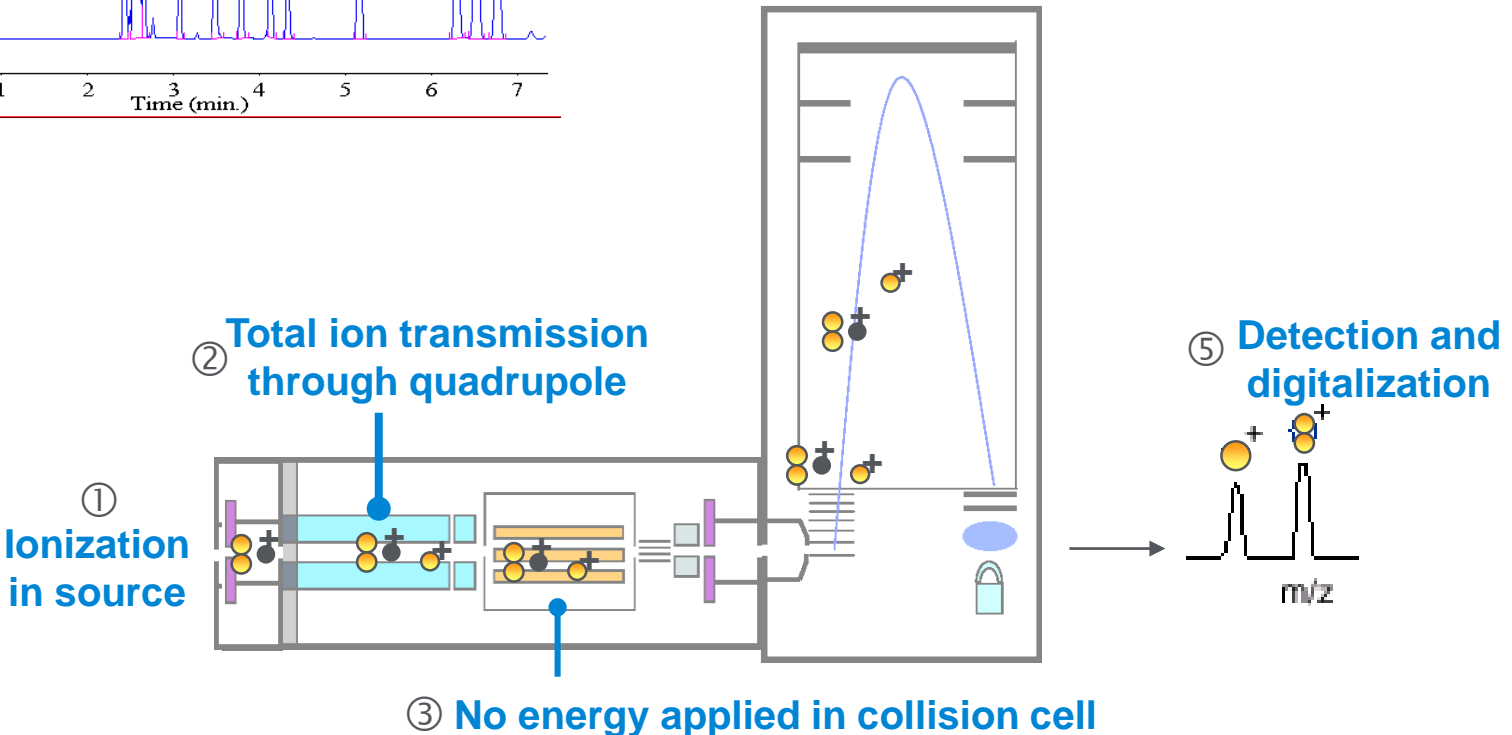# Q-TOF GC/MS: A typical spectrum in MS mode

Single Quad Scan Data
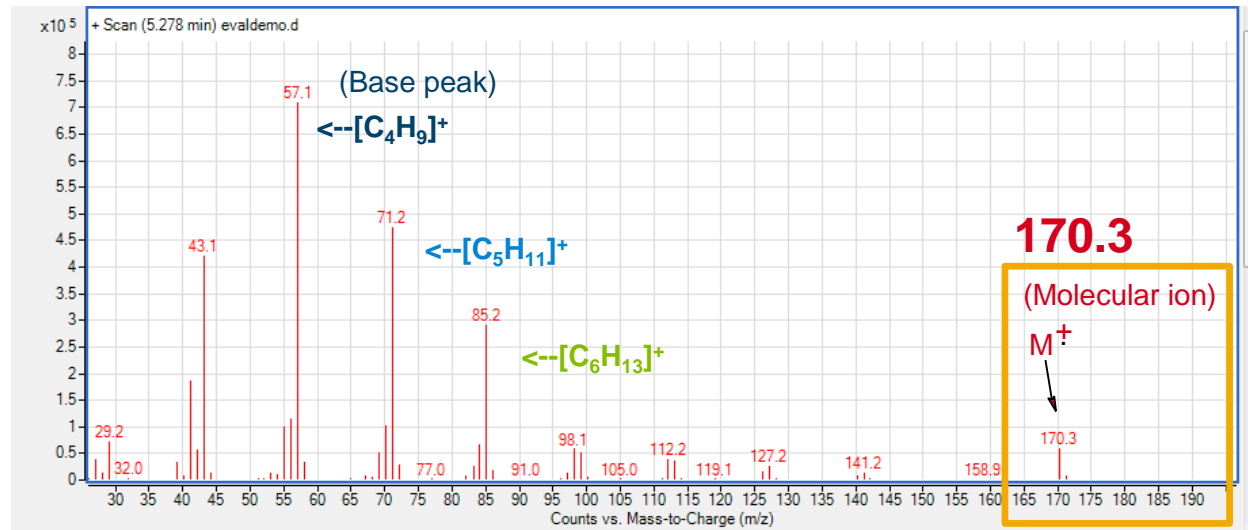
Q-TOF Data

The molecular ion in EI mode: loss of one electron, a radical cation, M+

Base peak: most abundant ion in spectrum.

Most fragment ions are even electron ions, except where rearrangements have occurred.

Single Quadrupole and TOF spectra will be slightly different due to the ion path length, TTI cut-off, etc.

Q-TOF GC/MS: Untargeted Screening and Identification of Unknowns by Q-TOF

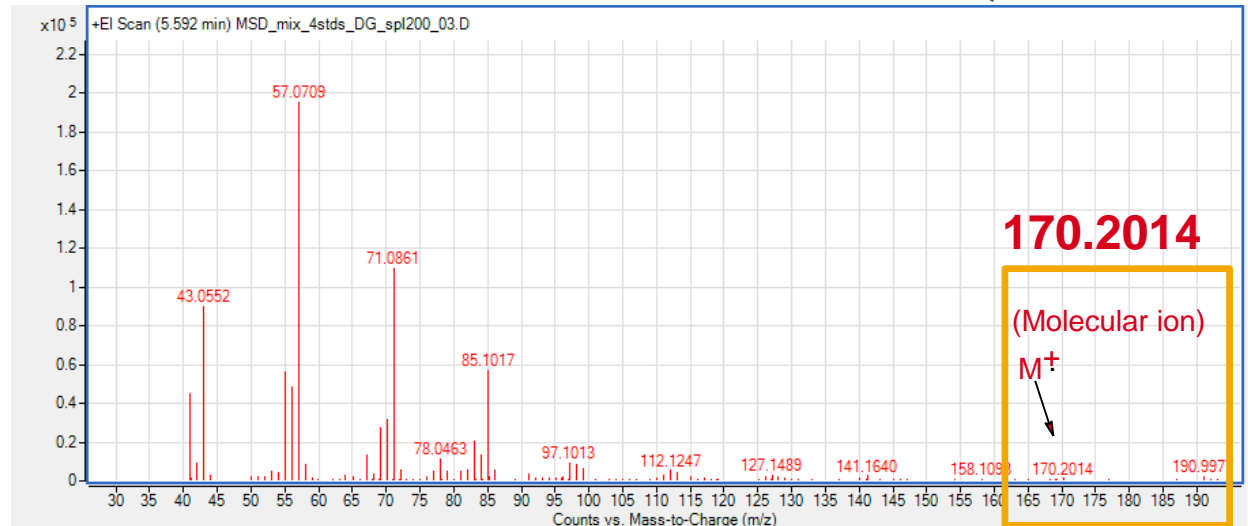**Broccoli sample**
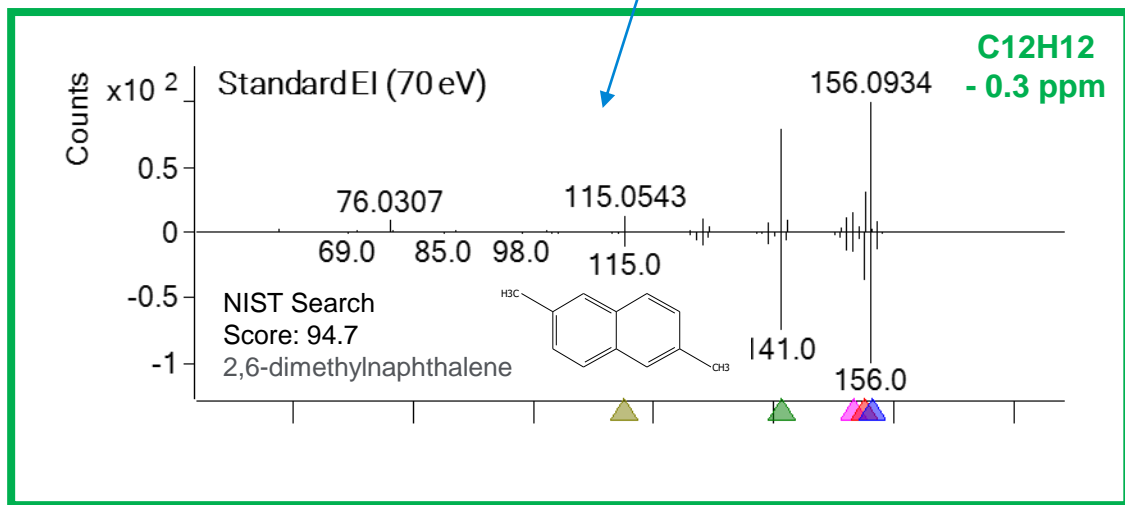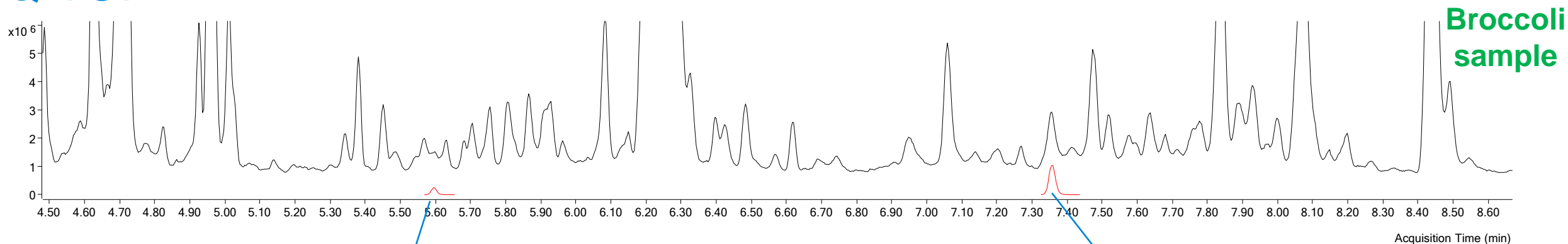
- Good library match and accurate mass confirm the identification
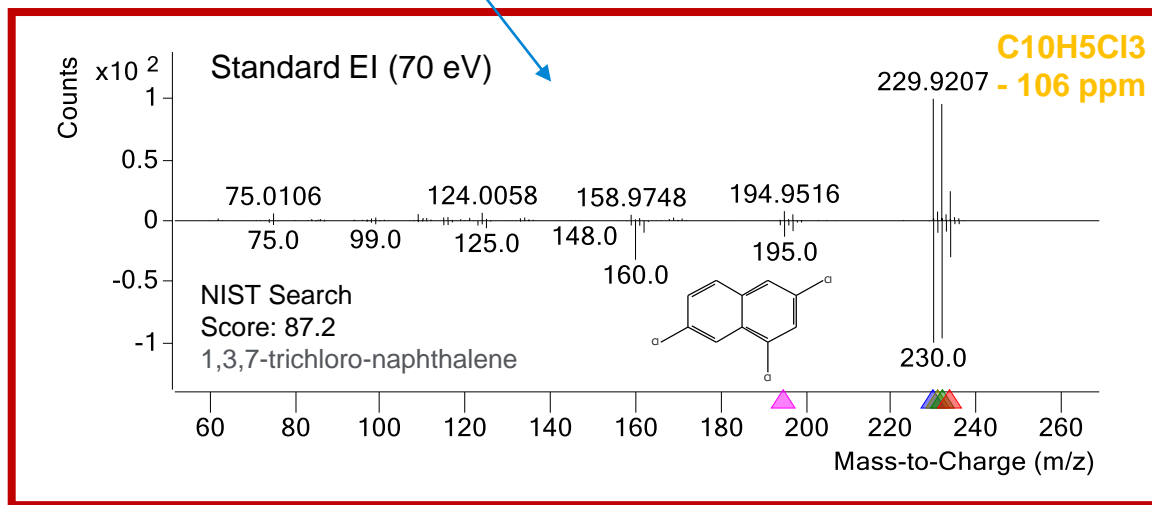
- Large mass error suggests different ID or unknown
- Accurate mass MS/MS help elucidate the candidate structure of true unknown

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Q-TOF GC/MS: Confirming structures with low energy EI MS/MS and accurate mass

Step 1: Confirm M+

Step 2: Confirm fragment ions
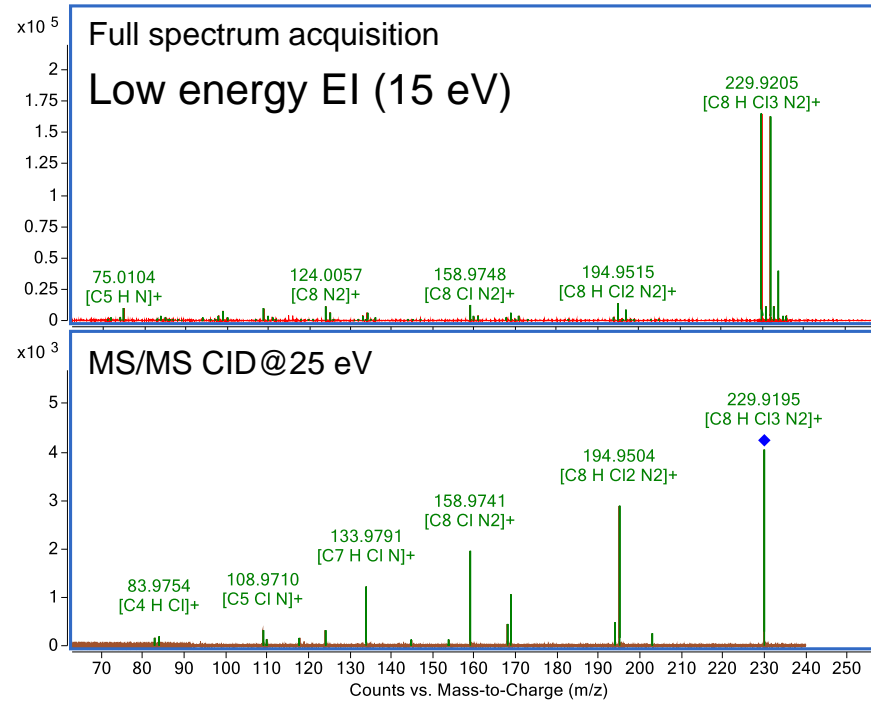
Step 3: Structure elucidation on candidate

Low energy EI (15 eV)
Isotope pattern and mass accuracy are good
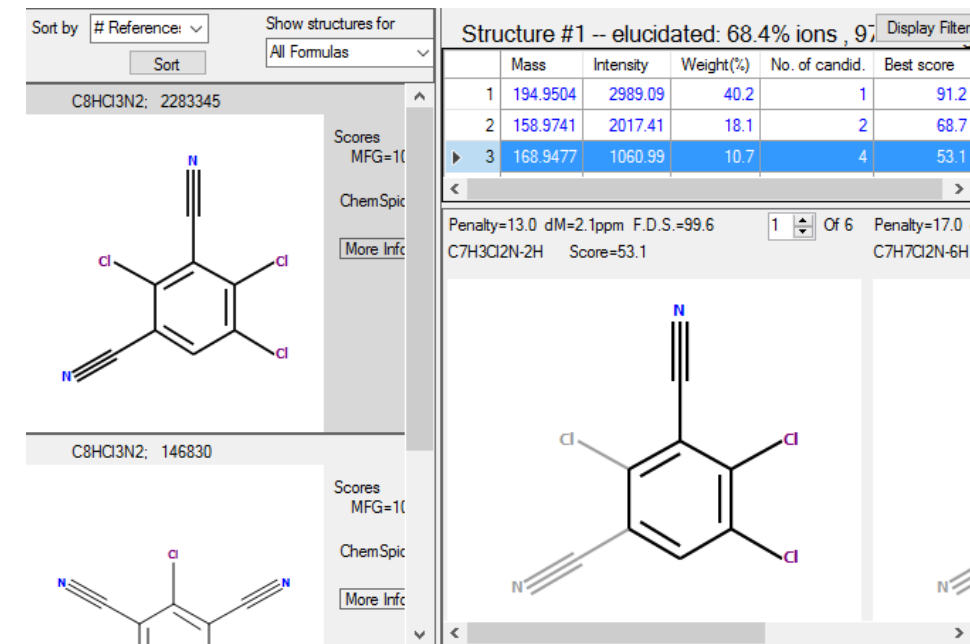
C8-H-Cl3-N2 turns out to be 2,4,5-Trichloroisophthalonitrile.
That is not NIST. Agilent Molecular Structure Correlator.
It is a degradation product of Chlorothalonil.

Q-TOF GC/MS: Summary

Newest GC/MS technique available from Agilent
- Provides, high-resolution, accurate-mass scan data with sensitive detection.
- High-resolution allows results in BOTH high selectivity over matrix AND sensitive detection
- High-resolution and accurate mass provides chemical formulas for spectral interpretation of true unknowns
- Easier method development than TQ
- Much higher selectivity than SQ but not as high as TQ

Modes of use
- Quantitative analysis (how much is in there?)
  - EICs using target ion and qualifiers
  - Since all data is scan mode, unlimited number of analytes
- Qualitative analysis (what is in there?)
  - Screening for large number (>1000) of pesticides using accurate mass fragments
  - Searching of spectra against libraries like NIST, Agilent RTL libraries, or others
  - Spectral interpretation of true unknowns using accurate mass data

Limitations
- High cost
- Bench space required
- Very large data file size, requires high powered data processing

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Analysis of Pesticides and Environmental Pollutants in Essential Oils Using Multi-Platform GC/MSD, GC/TQ and GC/Q-TOF

Kai Chen

Agilent Technologies, Inc., Santa Clara

Bruce Quimby

Agilent Technologies, Inc., Wilmington, Delaware

Vivian Xianyu Chen

Agilent Technologies (Shanghai) Co., Ltd., Shanghai, China

Trusted Answers

Using SQ, TQ, and Q-TOF to Examine Essential Oils for Pesticide Residue

GC/MSD: Use a retention time locked (RTL) spectral library of 950+ compounds and screen based on match scores of deconvoluted spectra.

GC/TQ: Initial screening of samples using scan mode (as above). Subsequent MRM analysis for trace level identity confirmation and quantitation.
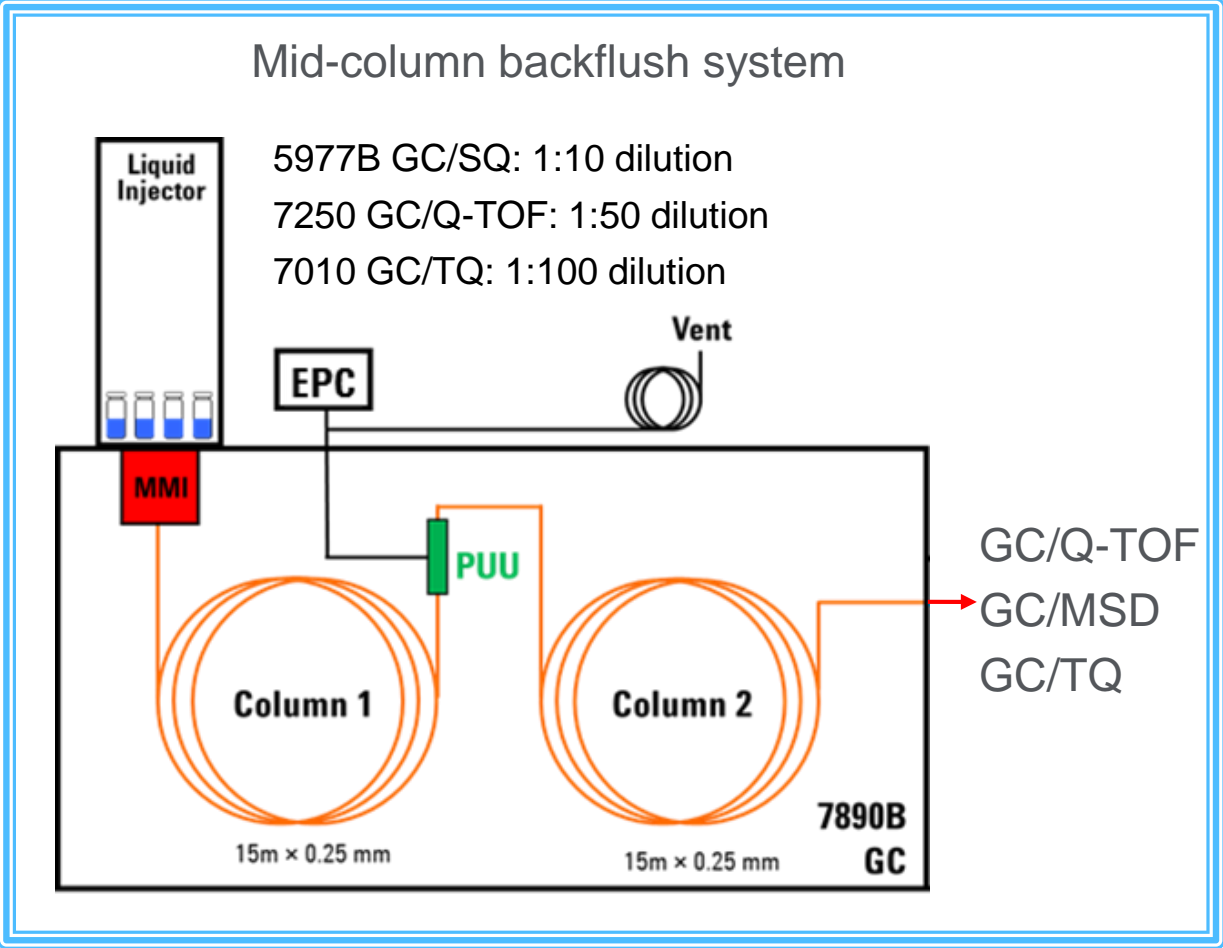
GC/Q-TOF: Use an RTL library containing accurate mass spectra for 850+ compounds and screen using six principle accurate mass ions, fragment ratios and RT matching.

This multi-platform GC/MSD, GC/TQ and GC/Q-TOF approach not only provides comprehensive and flexible analysis methods, but also offers a more streamlined workflow using different platforms.

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Samples and Data Acquisition

Essential oils as model complex matrices

- Grapefruit oil
- Lemon oil
- Neroli oil
- Orange oil (Brazil origin)
- Orange oil (California origin)
- …

Mid-column backflush system

5977B GC/SQ: 1:10 dilution

7250 GC/Q-TOF: 1:50 dilution

7010 GC/TQ: 1:100 dilution

GC/Q-TOF
GC/MSD
GC/TQ

Grapefruit Oil: Severe matrix interferences

GC/SQ TIC Grapefruit Oil
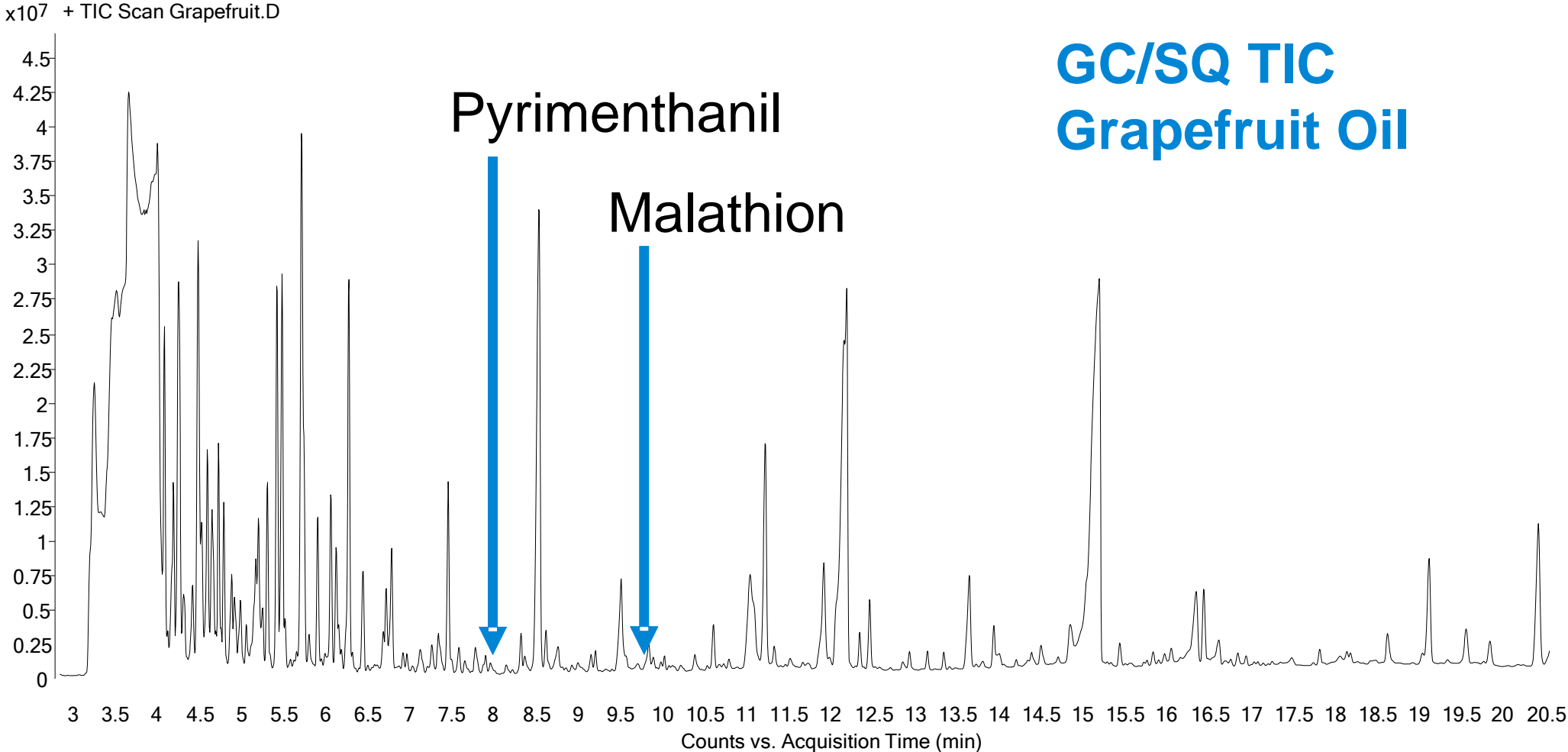
Pyrimenthanil Found In Screening By All Three Systems
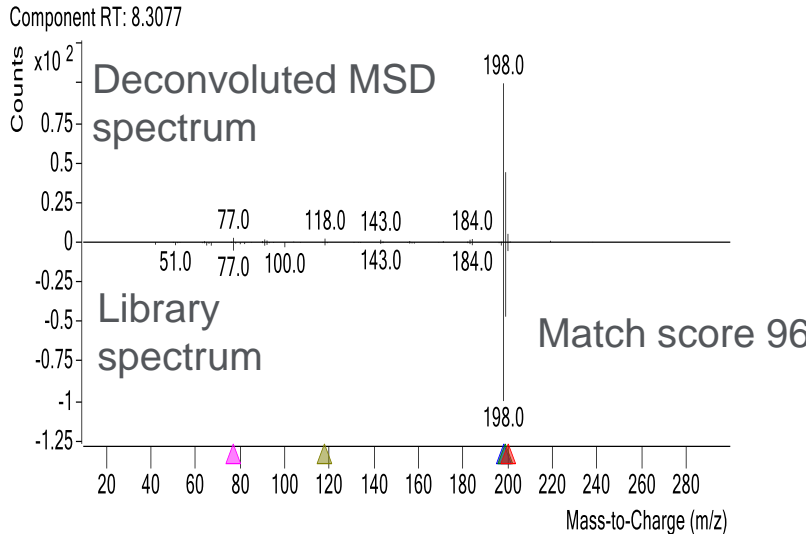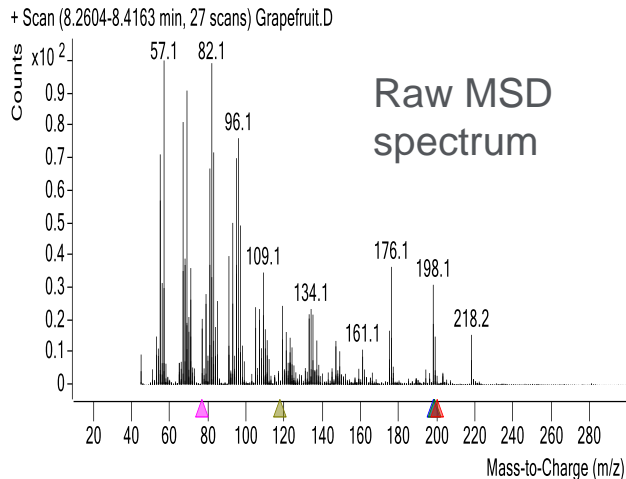
Quantitation with TQ MRM mode found Pyrimenthanil was 29.4 ng/mL as injected (100:1 dilution)

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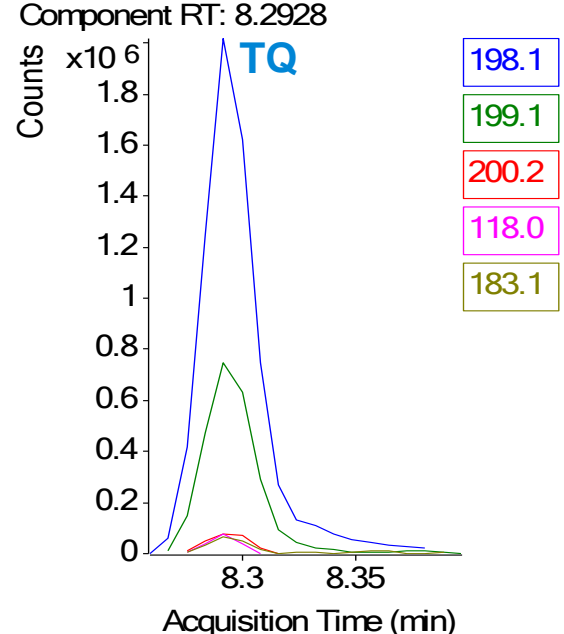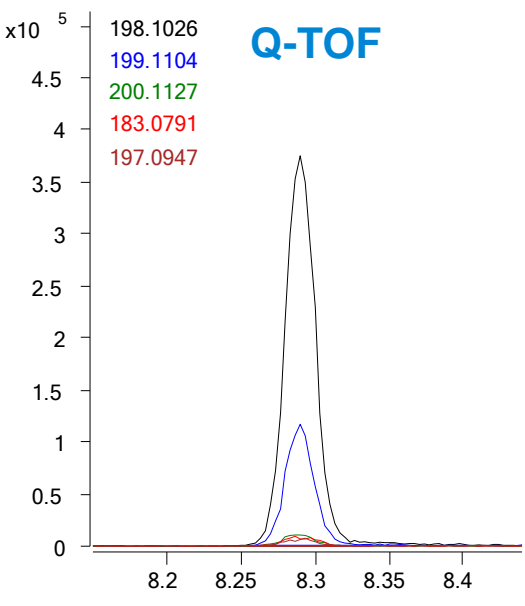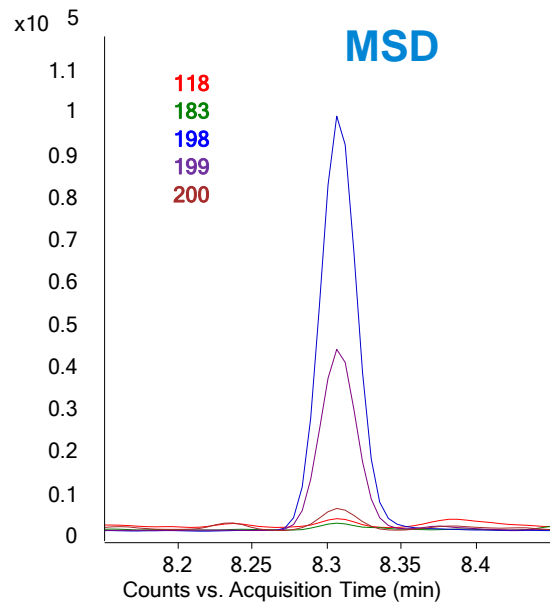
Malathion: Very Large Interferences

SQ is overwhelmed by interferences. No indication of Malathion.

Q-TOF finds Malathion with good RT match, fragment ratio score and mass accuracy.

TQ scan screen also missed Malathion, but MRM mode found it easily, with quant value = 1.26 ng/mL as injected (100:1 dilution).

Results Summary Table

Identification and quantification results from grapefruit oil by multiple platforms

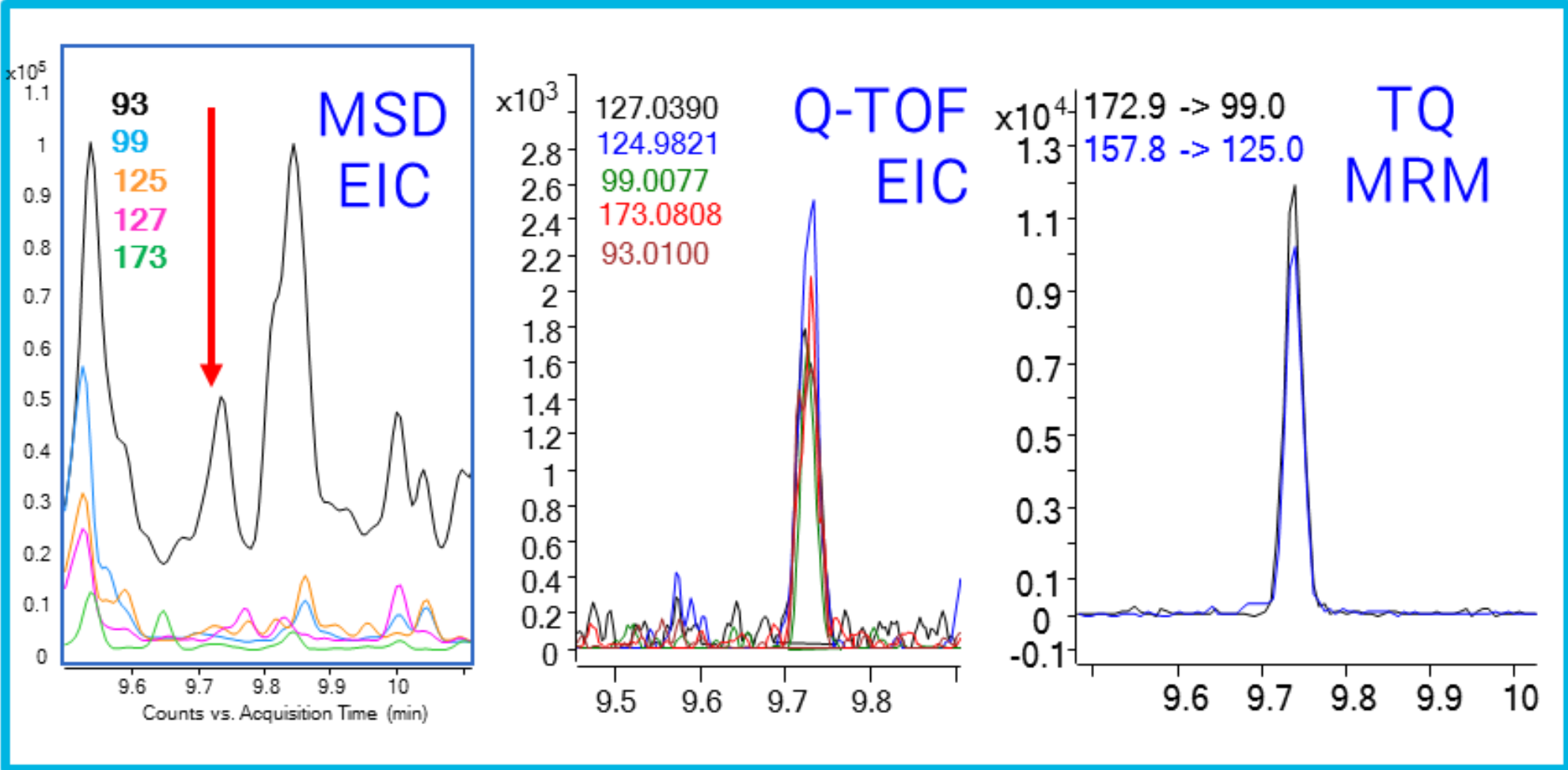| Compound Name	GC/MSD		GC/Q-TOF			GC/TQ (MRM)	
	Match Score	RT Diff (min)	Frag Ratio Score	RT Diff (min)	Mass Diff (ppm)	RT Diff (min)	Conc. (ng/mL)
Aniline	68	0.068	rejected after review			n.d.	n.d.
Pyrimethanil	96	0.023	98.05	0.015	3.68	0.012	29.41
Chlorpyrifos Methyl	79	0.000	99.92	0.005	1	0.006	3.33
Chlorpyrifos	93	-0.002	95.33	0.006	0.42	0.009	30.69
Imazalil	57	0.047	95.99	0.048	0.88	0.019	25.20
Pyriproxyfen	81	0.020	85.83	0.019	1.55	0.016	10.86
Malathion	n.d.	n.d.	82.15	0.014	0.17	0.011	1.26
Primiphos-methyl *	n.d.	n.d.	n.d.	n.d.	n.d.	0.011	0.22
Oxadixyl *	n.d.	n.d.	n.d.	n.d.	n.d.	0.021	0.36
Methidathion *	n.d.	n.d.	n.d.	n.d.	n.d.	0.004	0.54

* quantified by the routine multi-residue MRM method on TQ
n.d. = not detected or identified

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Summary

For discovery of pesticides present:

MSD or TQ Scan: Examine data file using:

- SureTarget spectral deconvolution and Agilent unit mass pesticide library.

Q-TOF Scan: Examine data file using:

- Accurate mass pesticides library (PCDL) with 850+ compounds via Find by Fragments in MassHunter Qualitative Analysis
- SureTarget spectral deconvolution and Agilent unit mass pesticide library in Unknowns Analysis for compounds not in PCDL

For quantitation of pesticides present:

MSD Scan or SIM: Quant using ions with least interference for discovered pesticides in that matrix.

TQ MRM: Quant using MRMs with least interference for discovered pesticides.

Q-TOF: Quant using accurate mass EICs with least interference for discovered pesticides.

Thank you for your attention!

Any questions?

Pesticides in Essential Oils

Essential oils are concentrated liquids containing volatile aroma compounds from various plants and are widely used in flavors and fragrances. Traces of pesticides are sometimes present in these oils, resulting in the need to screen for them.

This analysis is challenging due to the large background interference presented by oil matrices. Traditional GC/MS is limited in capacity to perform the task. This work demonstrates the use of advanced techniques for pesticide screening in essential oils.

Chromatographic Parameters

GC and MS Conditions	Value
Columns (2 ea.)	Agilent J&W HP-5ms UI, 15 m × 0.25 mm × 0.25 µm (P/N: 19091S-431 UI)
Inlet	MMI, 4 mm UI liner with wool
Injection	1 or 2 µL cold splitless
Carrier gas	Helium
Inlet flow (column 1)	~ 1 mL/min
PUU flow (column 2)	column 1 flow + 0.2 mL/min
Oven program	60°C for 1 min 40°C/min to 170°C for 0 min 10°C/min to 310°C for 3 min (Total run time: 20.75 min)
Backflush Conditions	5 min (postrun). 310 °C (oven). 50 psi (aux EPC pressure). 2 psi (inlet pressure)
Transfer line temperature	280 °C
Ion source temperature	280 °C
Quadrupole temperature	150 °C