



EURL-FV

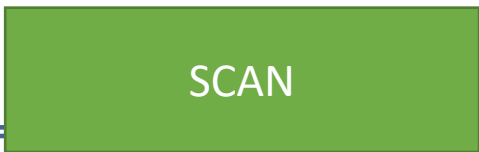
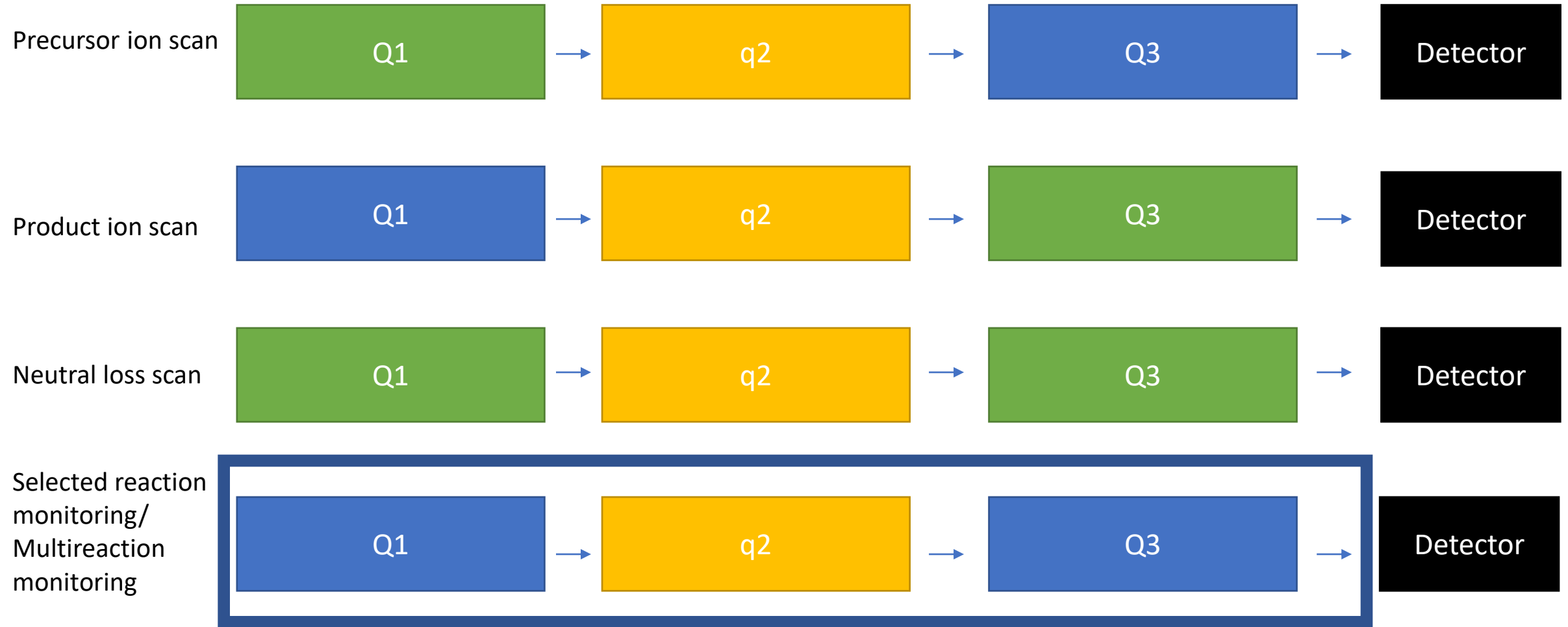


New advances in HRMS

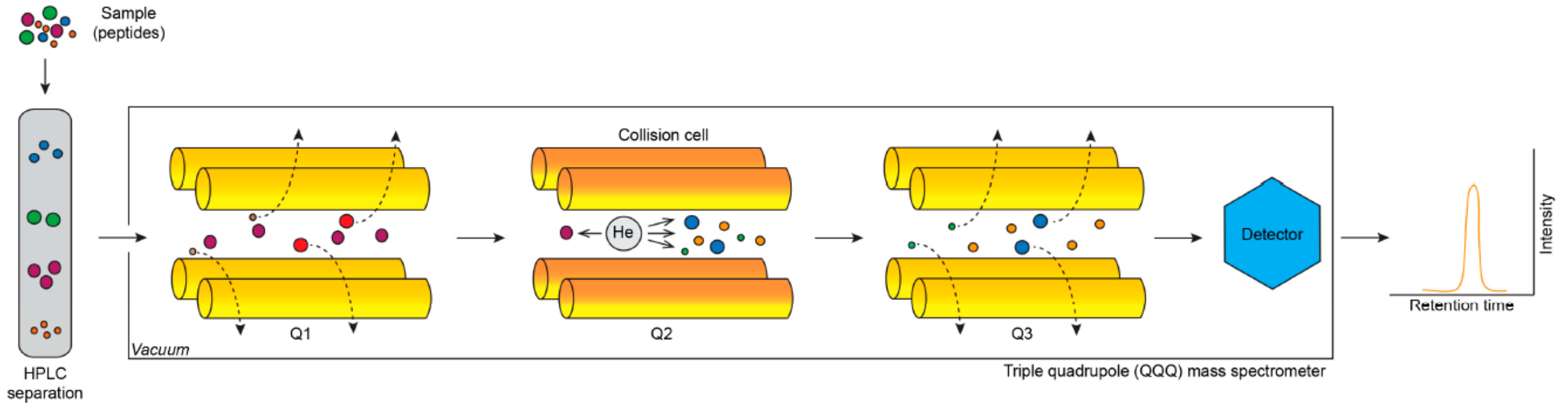
Łukasz Rajski

Practical Training - New Advances in MRM (on-line training)
21-22/01/2021

Triple quadrupole



Triple quadrupole



Int. J. Mol. Sci. **2017**, *18*(12), 2697; <https://doi.org/10.3390/ijms18122697>

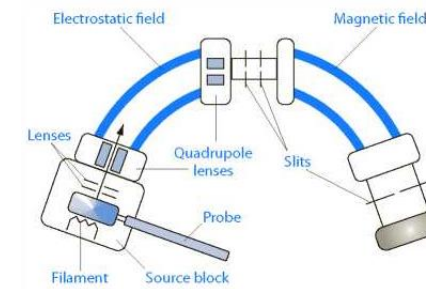
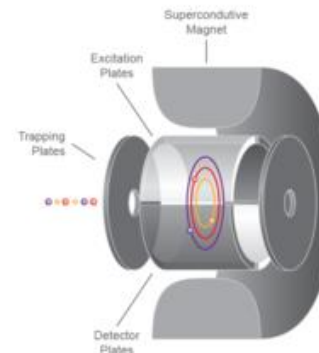
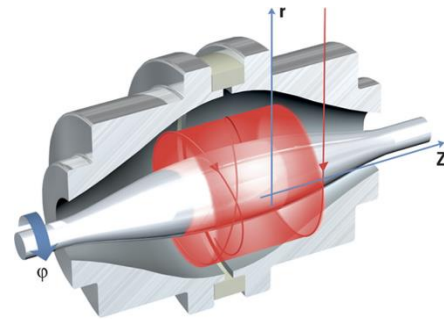
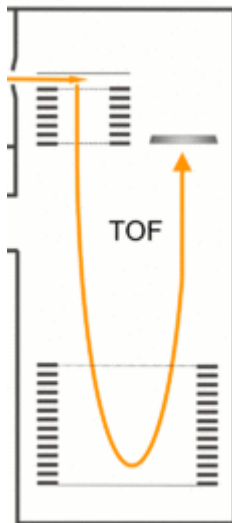
High-resolution mass spectrometry

Instrumentation:

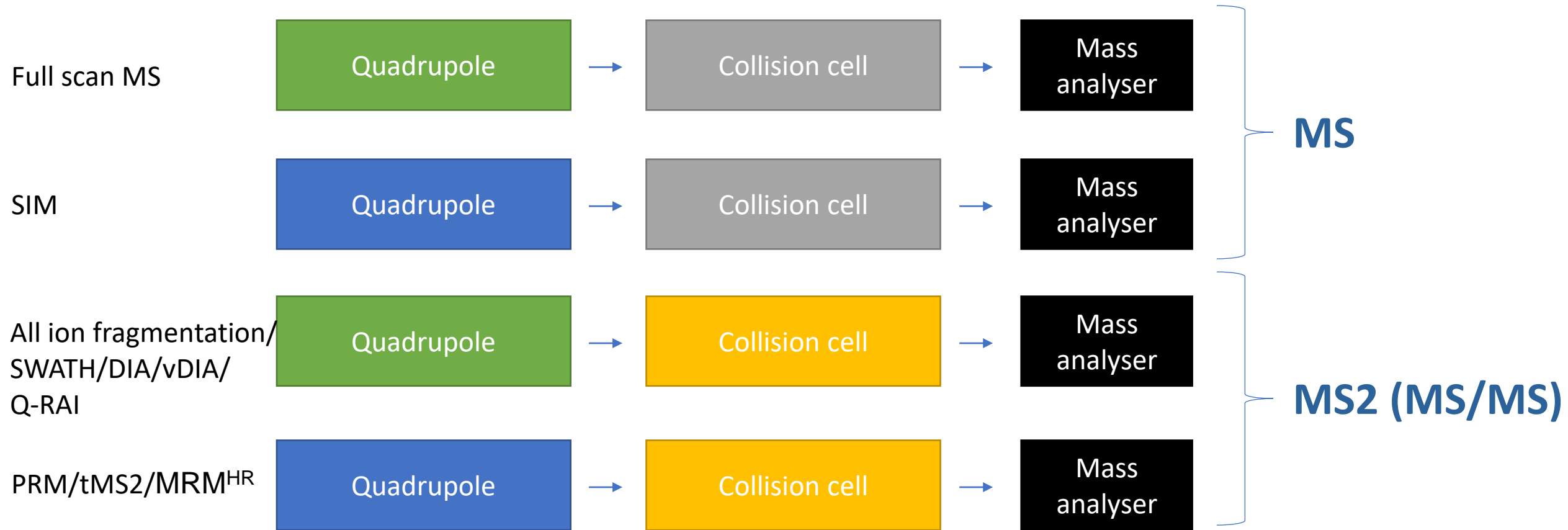
- Time of flight
- Orbitrap



QToF
QOrbitrap



High-resolution mass spectrometry



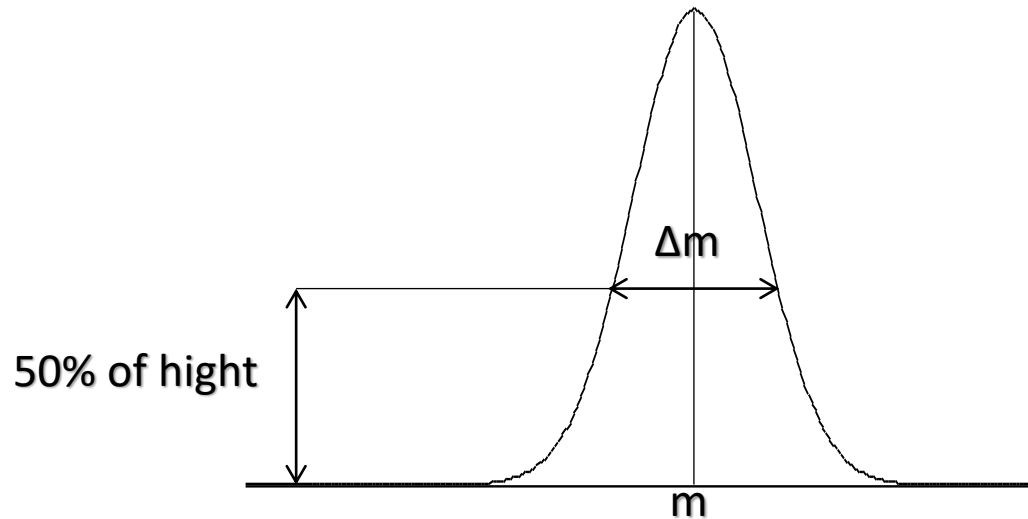
High-resolution mass spectrometry

Resolution in mass spectroscopy

1. (energy): By analogy with the peak width definition for mass resolution, a peak showing the number of ions as a function of their translational energy should be used to give a value for the energy resolution.
 2. (10 per cent valley definition): Let two peaks of equal height in a mass spectrum at masses m and $m - \Delta m$ be separated by a valley which at its lowest point is just 10 per cent of the height of either peak. For similar peaks at a mass exceeding m , let the height of the valley at its lowest point be more (by any amount) than ten per cent of either peak height. Then the resolution (10 per cent valley definition) is $m/\Delta m$. It is usually a function of m . The ratio $m/\Delta m$ should be given for a number of values of m .
3. (peak width definition): For a single peak made up of singly charged ions at mass m in a mass spectrum, the resolution may be expressed as $m/\Delta m$ where Δm is the width of the peak at a height which is a specified fraction of the maximum peak height. It is recommended that one of three values 50%, 5% or 0.5% should always be used. For an isolated symmetrical peak recorded with a system which is linear in the range between 5% and 10% levels of the peak, the 5% peak width definition is technically equivalent to the 10% valley definition. A common standard is the definition of resolution based upon Δm being Full Width of the peak at Half its Maximum height, sometimes abbreviated 'FWHM'. This acronym should preferably be defined the first time it is used.

High-resolution mass spectrometry

Resolution at FWHM - full-width half maximum



$$R = \frac{m}{\Delta m}$$

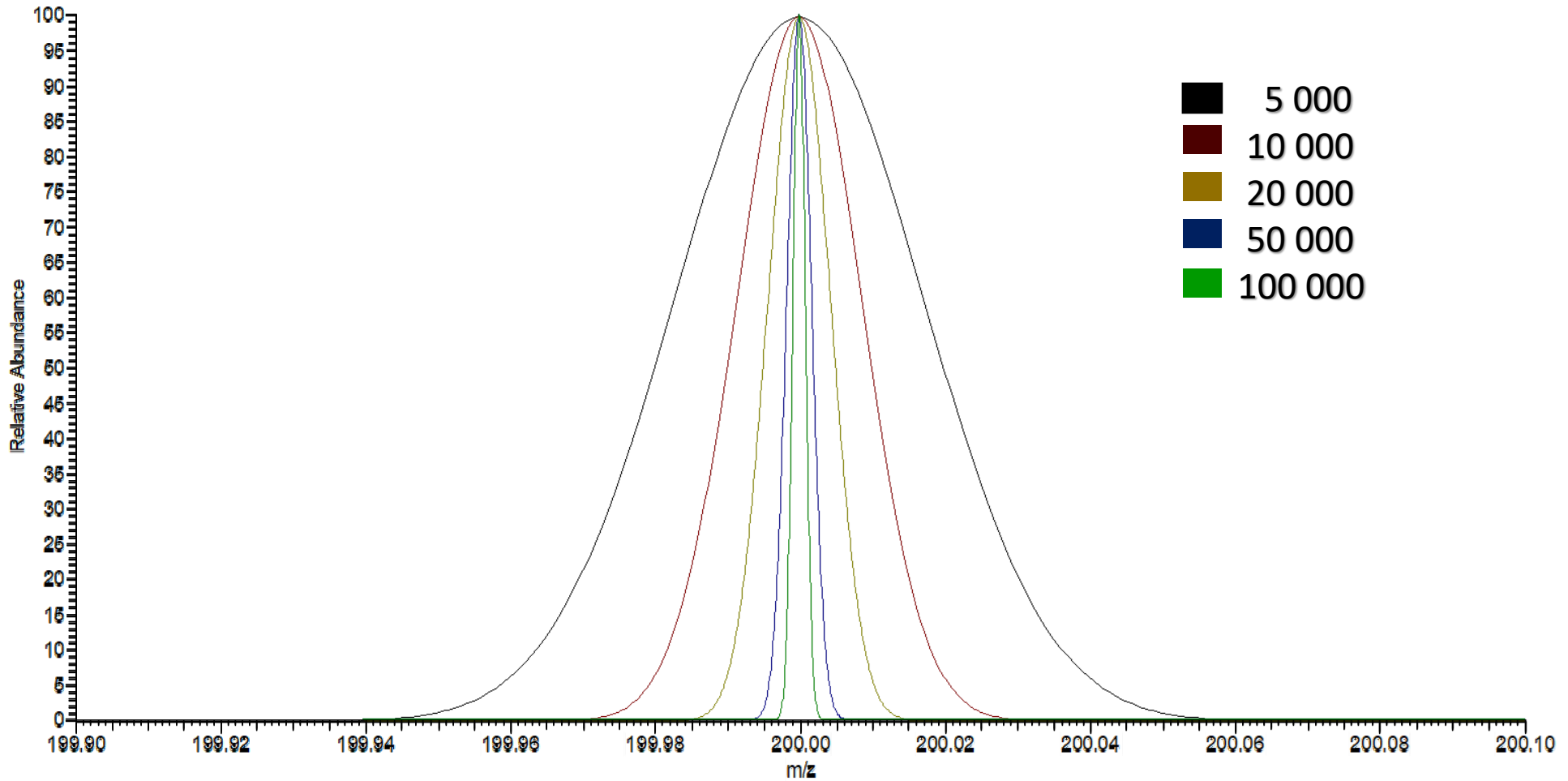
For example:

$$\frac{200.0000}{0.04} = 5\ 000$$

$$\frac{200.0000}{0.004} = 50\ 000$$

High-resolution mass spectrometry

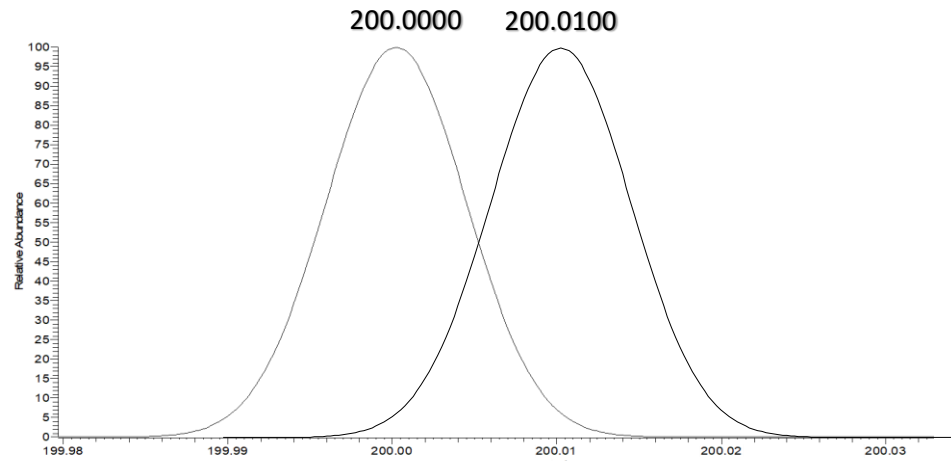
Effects of increased resolution



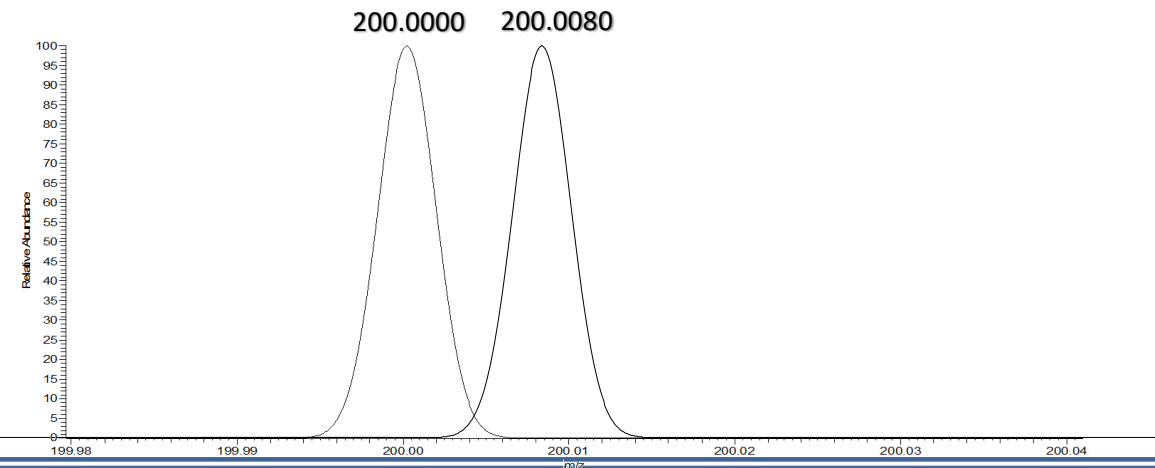
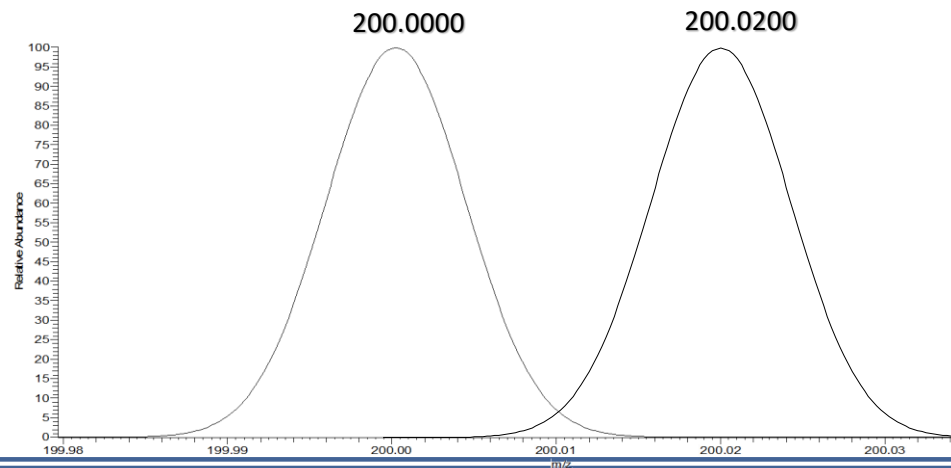
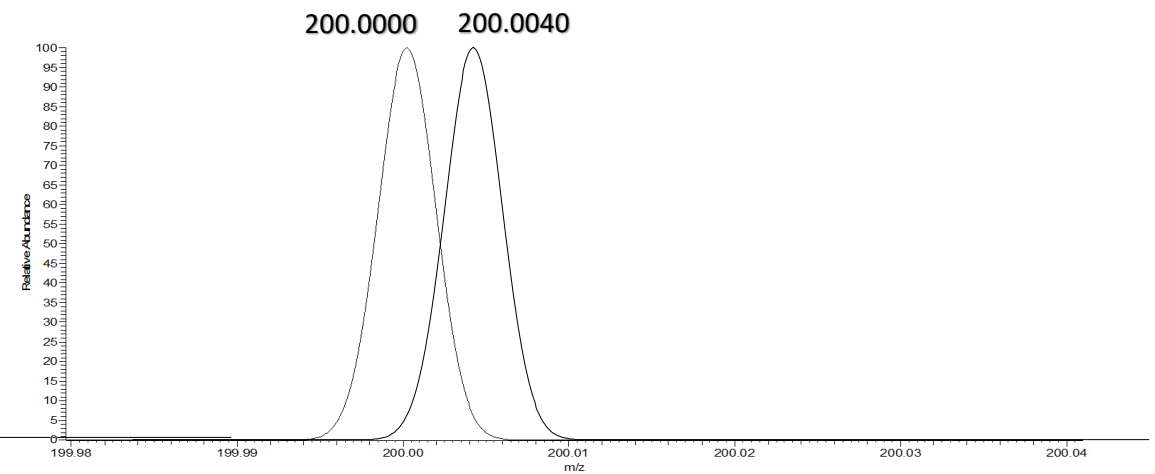
High-resolution mass spectrometry

Effects of increased resolution

Resolution: 20 000



Resolution: 50 000



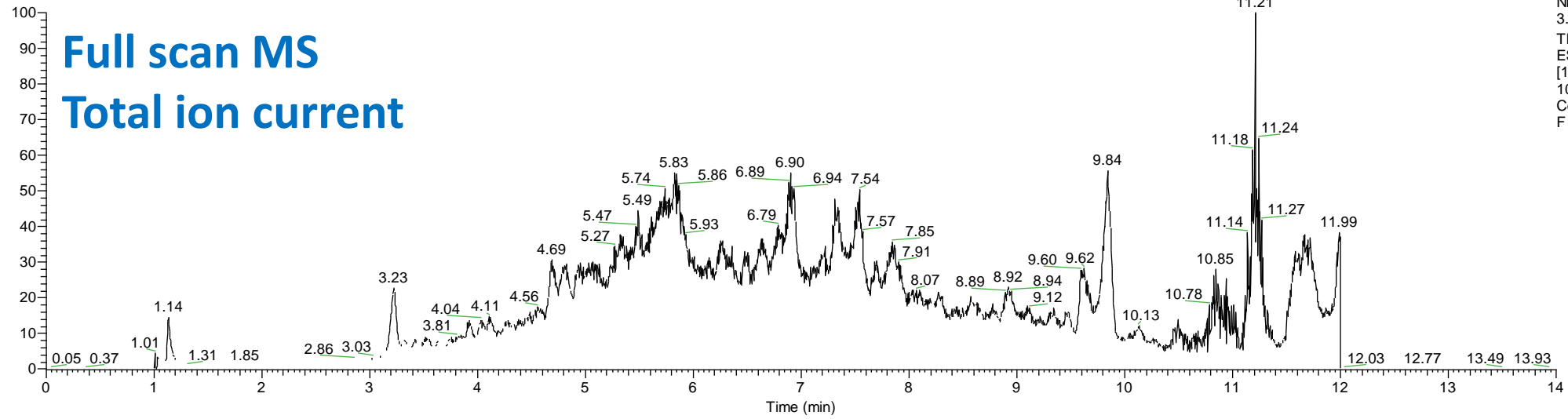
Onion

High-resolution mass spectrometry

D:\data\...\Cebolla_10ppb_AIF

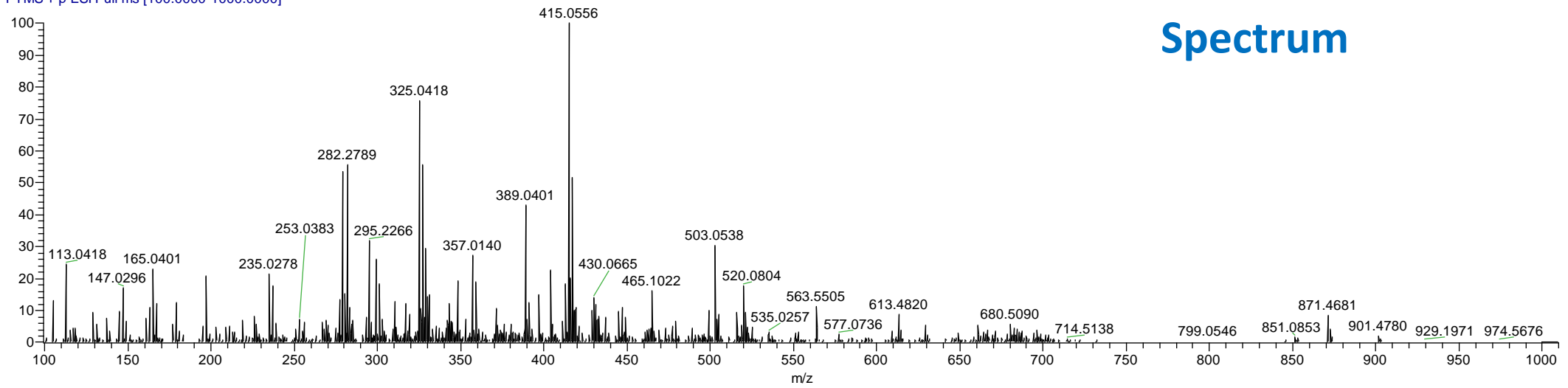
10/17/20 15:48:34

RT: 0.00 - 14.00



NL:
3.20E9
TIC F: FTMS + p
ESI Full ms
[100.0000-
1000.0000] MS
Cebolla_10ppb_AI
F

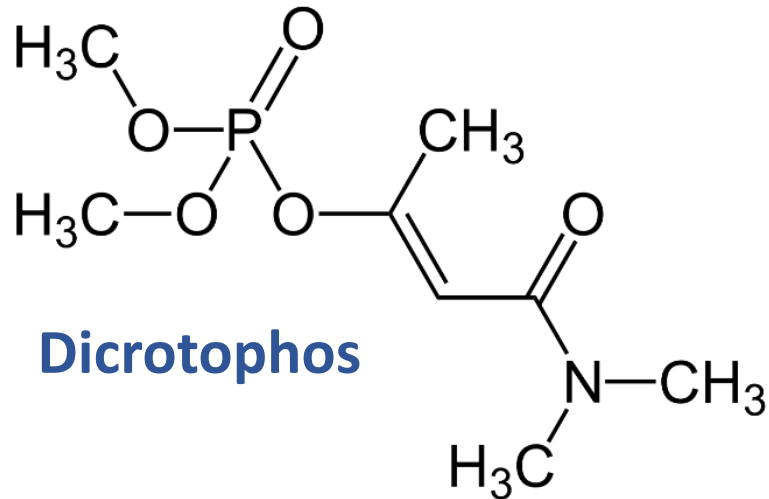
Cebolla_10ppb_AIF #7-6147 RT: 0.02-14.00 AV: 3071 NL: 1.07E7
T: FTMS + p ESI Full ms [100.0000-1000.0000]



Nominal mass: mass of a molecular ion or molecule calculated using the isotope mass of the most abundant constituent element isotope of each element rounded to the nearest integer value and multiplied by the number of atoms of each element.

Exact mass: calculated mass of an ion or molecule with specified isotopic composition.

High-resolution mass spectrometry



C	8 · 12	= 96
H	17 · 1.00783	= 17.13311
N	1 · 14.0031	= 14.0031
O	5 · 15.9949	= 79.9745
P	1 · 30.9738	= 30.9738
		<hr/>
		238.0845
		- 0.000549
		<hr/>
		238.0840

High-resolution mass spectrometry

Mass error:

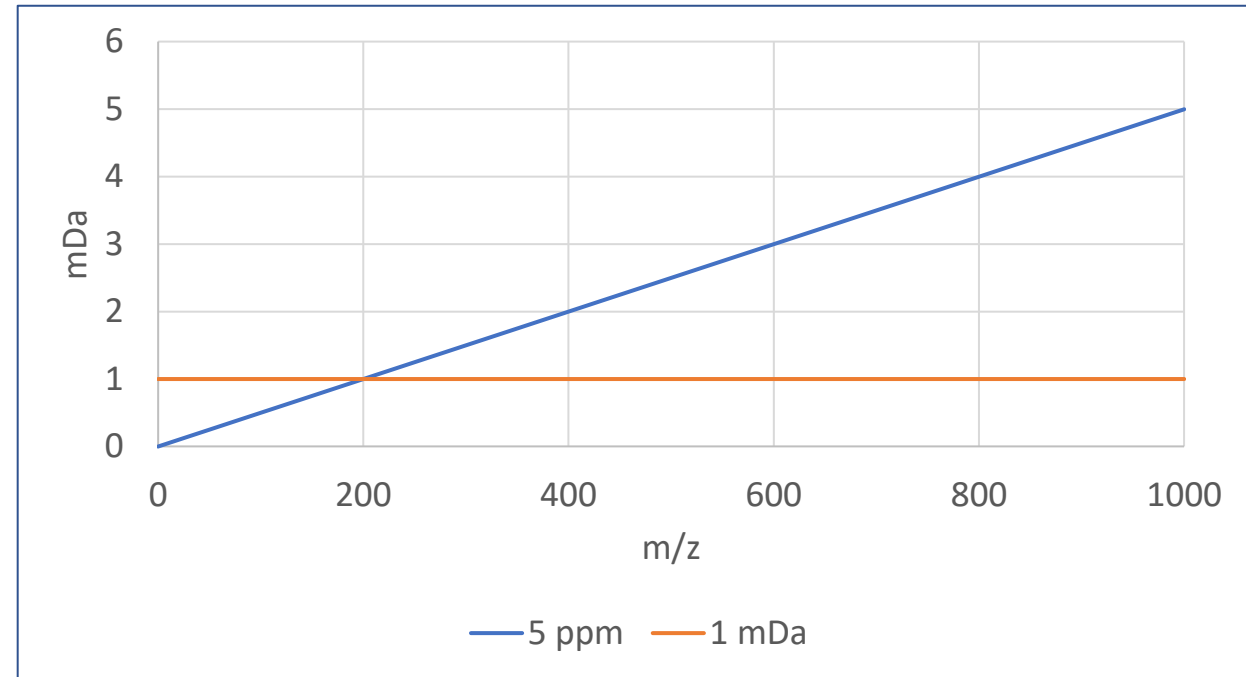
- absolute (expressed in mDa)
- relative (expressed in ppm)

$$5\text{ppm} = 0.000005 \text{ Da} = 0.005 \text{ mDa}$$

$$m/z \ 100 \cdot 0.000005 = 0.0005 \text{ Da}$$

$$m/z \ 200 \cdot 0.000005 = 0.001 \text{ Da}$$

$$m/z \ 400 \cdot 0.000005 = 0.002 \text{ Da}$$



$$m/z \ 100.0000 \pm 5 \text{ ppm} = m/z \ 99.9995 - 100.0005$$

$$m/z \ 200.0000 \pm 5 \text{ ppm} = m/z \ 199.9990 - 200.0010$$

$$m/z \ 400.0000 \pm 5 \text{ ppm} = m/z \ 399.9980 - 400.0020$$

$$m/z \ 100.0000 \pm 1 \text{ mDa} = m/z \ 99.9990 - 100.0010$$

$$m/z \ 200.0000 \pm 1 \text{ mDa} = m/z \ 199.9990 - 200.0010$$

$$m/z \ 400.0000 \pm 1 \text{ mDa} = m/z \ 399.9990 - 400.0010$$

Onion

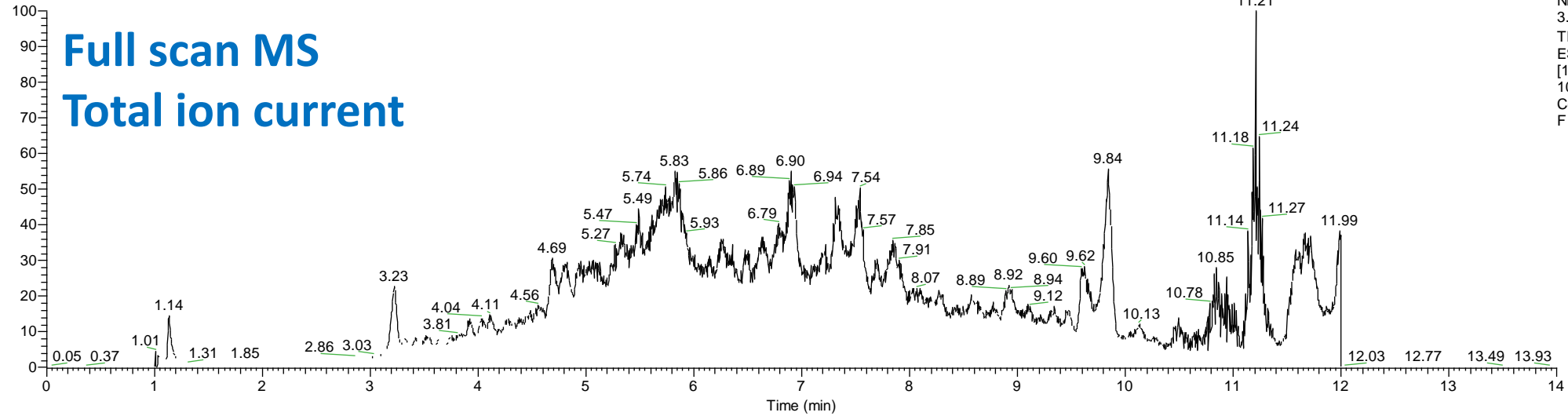
High-resolution mass spectrometry

D:\data\...\Cebolla_10ppb_AIF

10/17/20 15:48:34

RT: 0.00 - 14.00

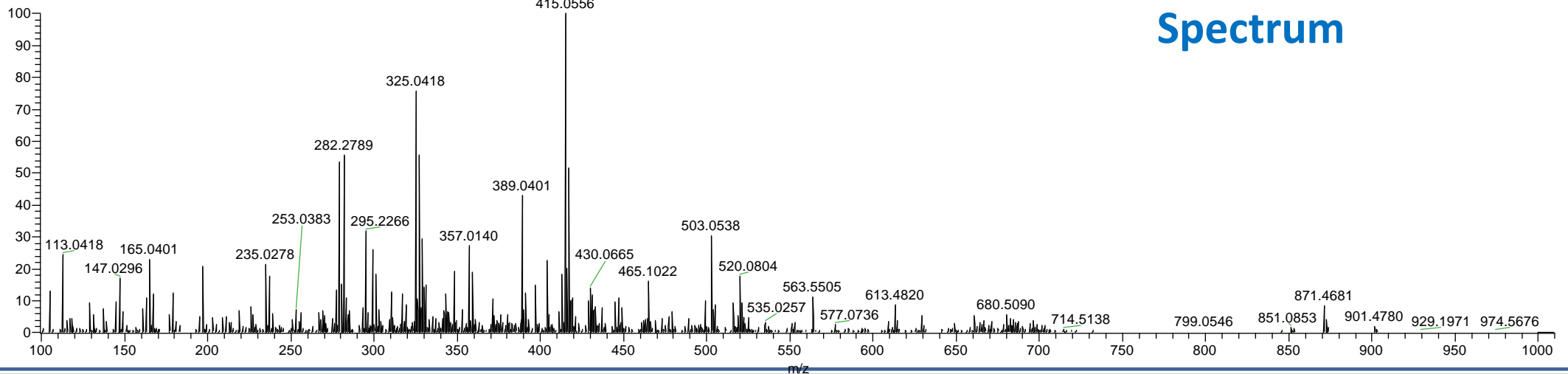
Full scan MS Total ion current



NL:
3.20E9
TIC F: FTMS + p
ESI Full ms
[100.0000-
1000.0000] MS
Cebolla_10ppb_AI
F

Cebolla_10ppb_AIF #7-6147 RT: 0.02-14.00 AV: 3071 NL: 1.07E7
T: FTMS + p ESI Full ms [100.0000-1000.0000]

Spectrum



Onion

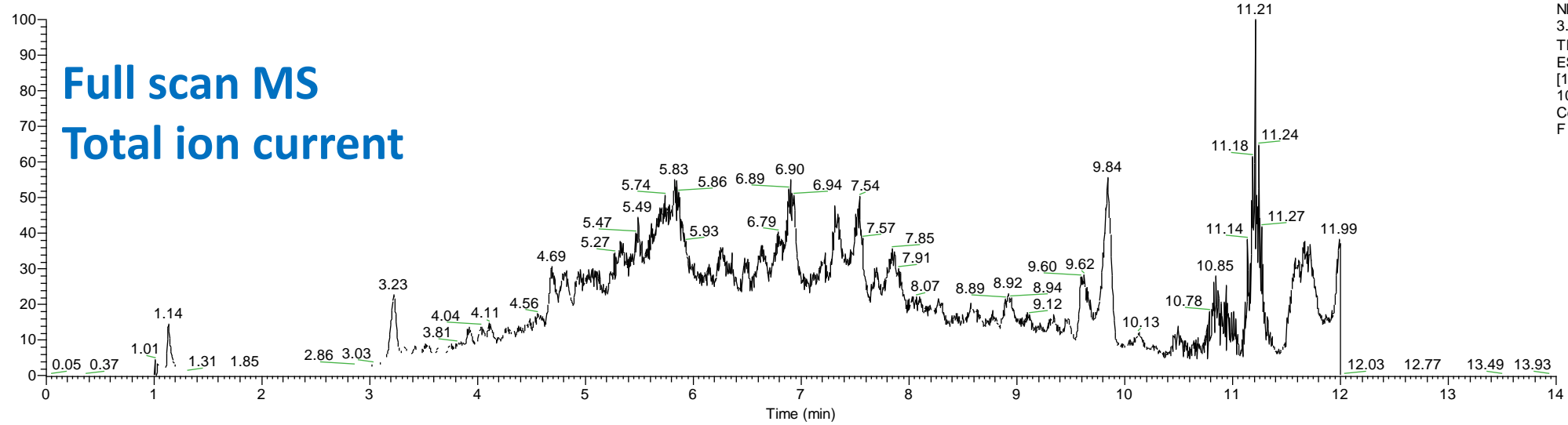
High-resolution mass spectrometry

D:\data...\Cebolla_10ppb_AIF

10/17/20 15:48:34

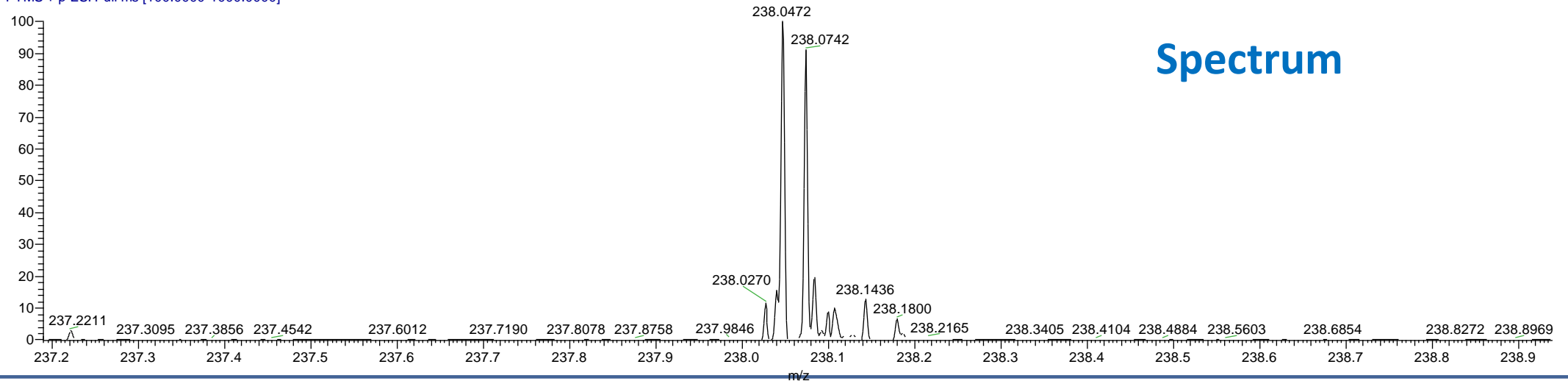
RT: 0.00 - 14.00

Full scan MS
Total ion current



NL:
 3.20E9
 TIC F: FTMS + p
 ESI Full ms
 [100.0000-
 1000.0000] MS
 Cebolla_10ppb_AI
 F

Cebolla_10ppb_AIF #7-6147 RT: 0.02-14.00 AV: 3071 NL: 1.39E5
 T: FTMS + p ESI Full ms [100.0000-1000.0000]



Spectrum

Onion

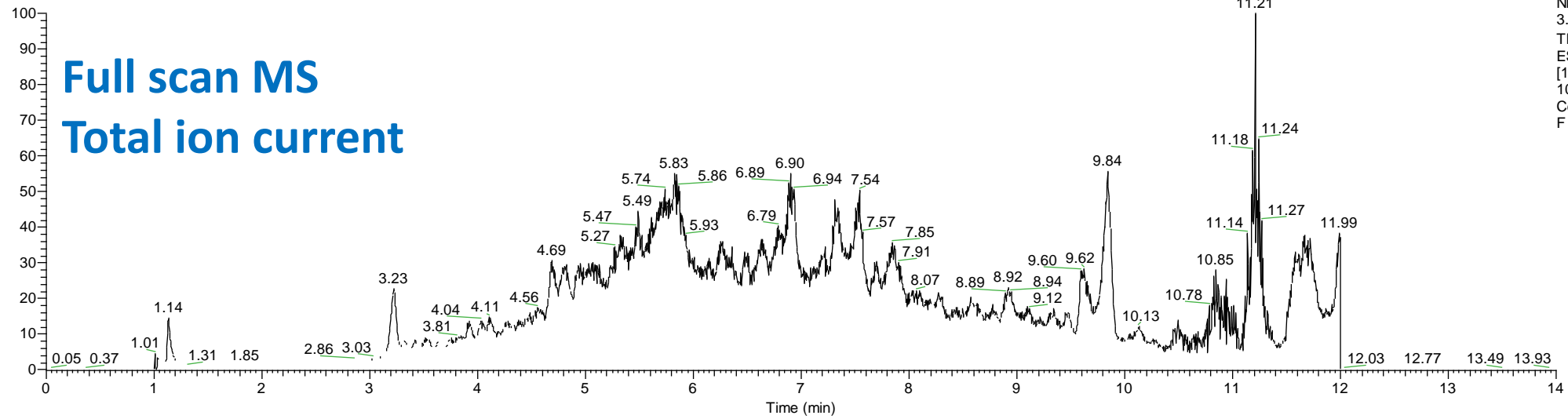
High-resolution mass spectrometry

D:\data...\Cebolla_10ppb_AIF

10/17/20 15:48:34

RT: 0.00 - 14.00

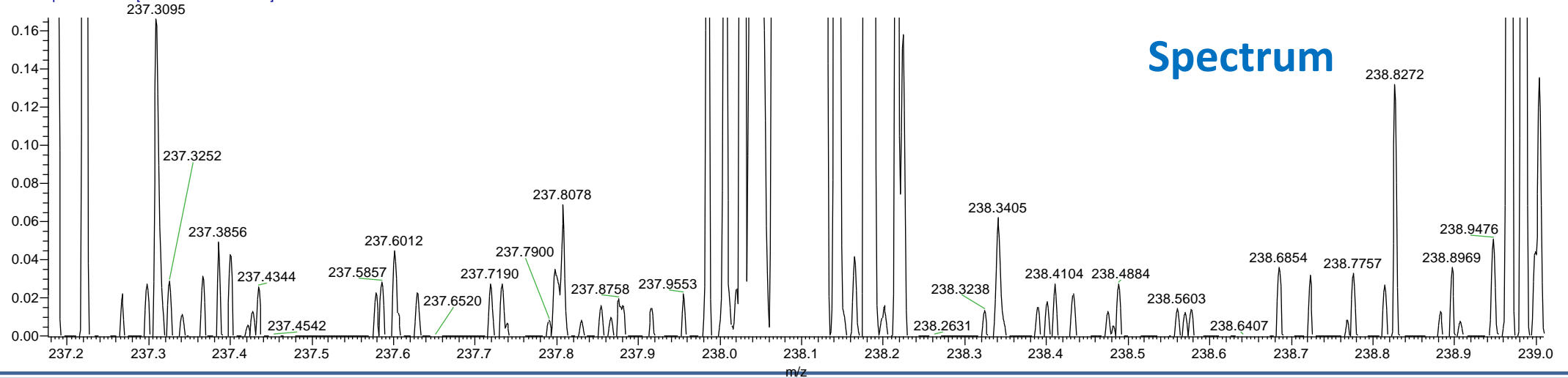
Full scan MS
Total ion current



NL:
3.20E9
TIC F: FTMS + p
ESI Full ms
[100.0000-
1000.0000] MS
Cebolla_10ppb_AI
F

Cebolla_10ppb_AIF #7-6147 RT: 0.02-14.00 AV: 3071 NL: 1.39E5
T: FTMS + p ESI Full ms [100.0000-1000.0000]

Spectrum



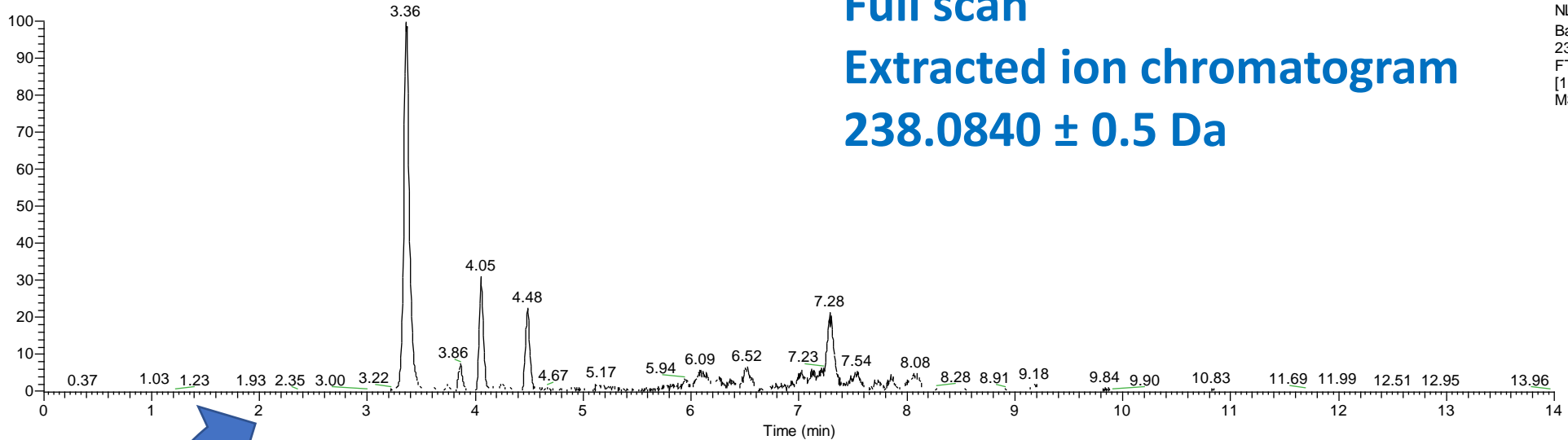
Onion

High-resolution mass spectrometry

D:\data...\Cebolla_10ppb_AIF

10/17/20 15:48:34

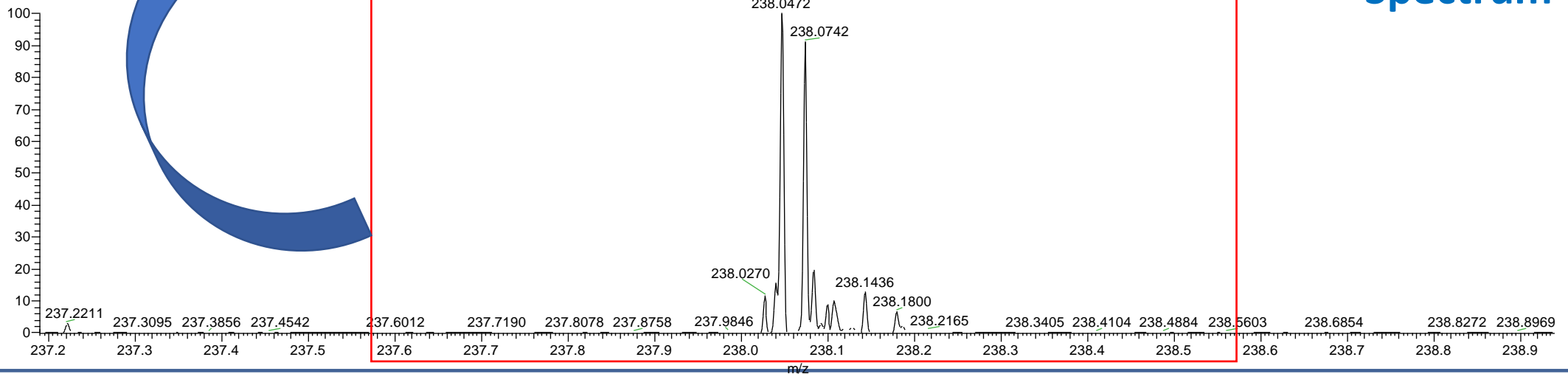
RT: 0.00 - 14.00



NL: 2.86E7
Base Peak m/z=
237.58390-238.58390 F:
FTMS + p ESI Full ms
[100.0000-1000.0000]
MS Cebolla_10ppb_AIF

Cebolla_10ppb_AIF #7-614... 2-14.00 AV: 3071 NL: 1.39E5
T: FTMS + p ESI Full ms [100.0000-1000.0000]

Spectrum



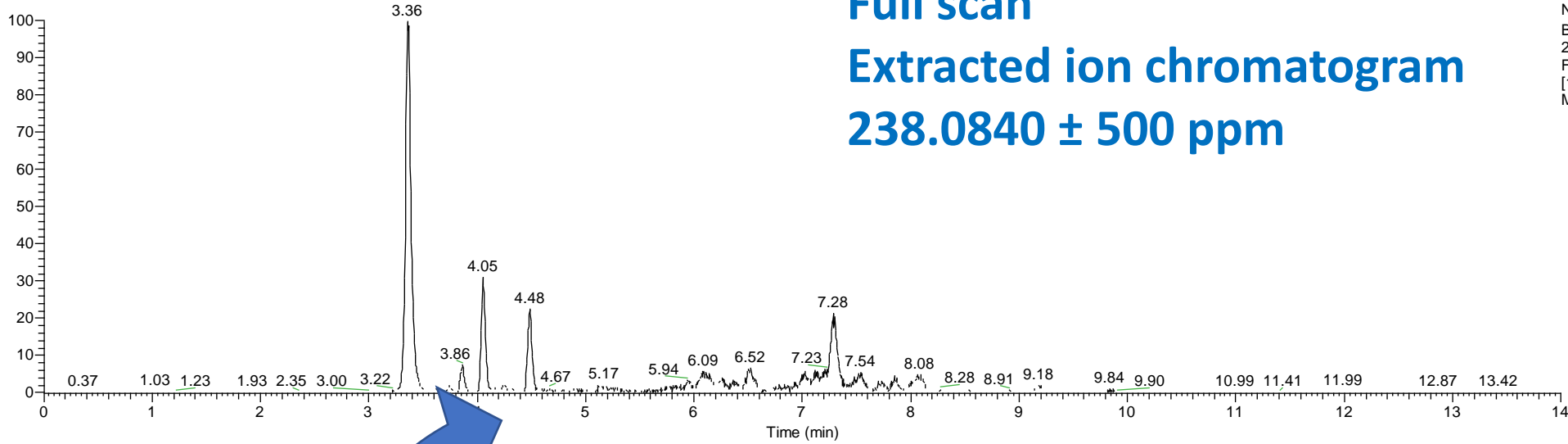
Onion

High-resolution mass spectrometry

D:\data\...\Cebolla_10ppb_AIF

10/17/20 15:48:34

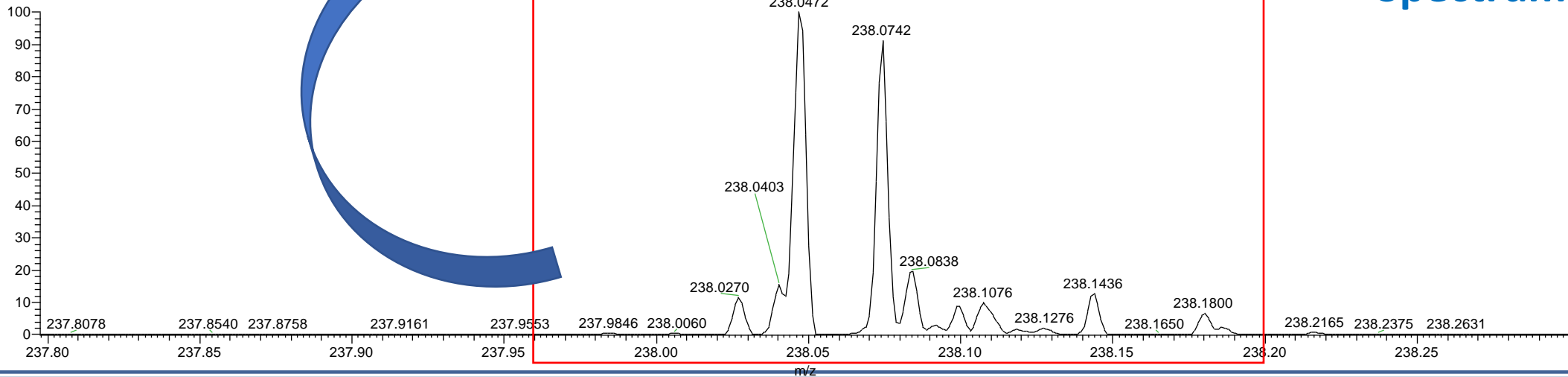
RT: 0.00 - 14.00



Full scan
Extracted ion chromatogram
238.0840 ± 500 ppm

NL: 2.86E7
Base Peak m/z=
237.96486-238.20294 F:
FTMS + p ESI Full ms
[100.0000-1000.0000]
MS Cebolla_10ppb_AIF

Cebolla_10ppb_AIF #7-6147 RT: 0.02-14.00 AV: 3071
T: FTMS + p ESI Full ms [100.0000-1000.0000]



Spectrum

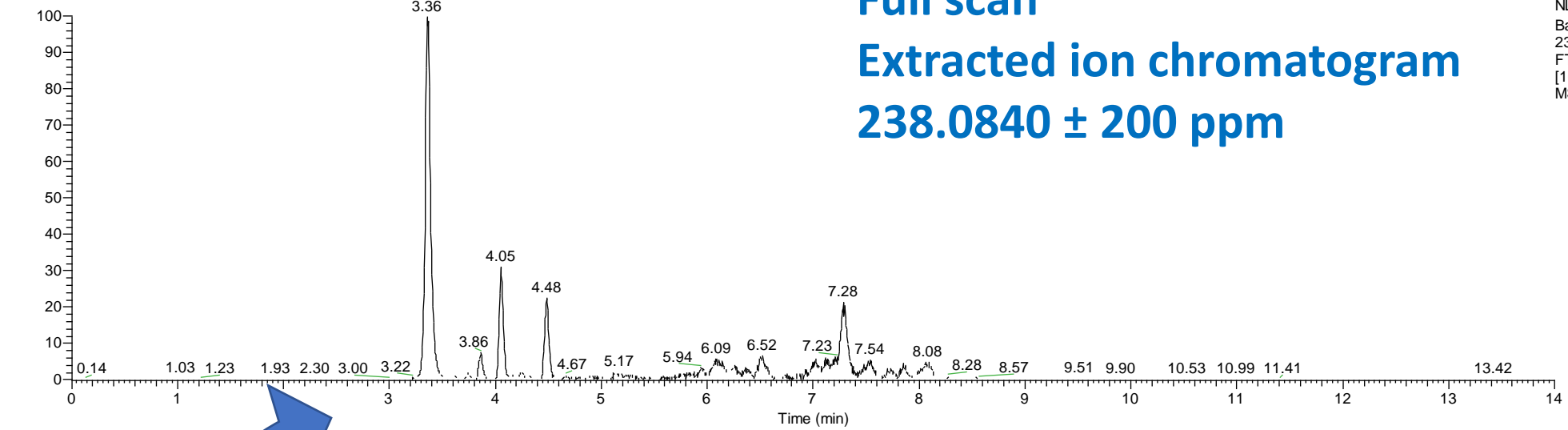
Onion

High-resolution mass spectrometry

D:\data...\Cebolla_10ppb_AIF

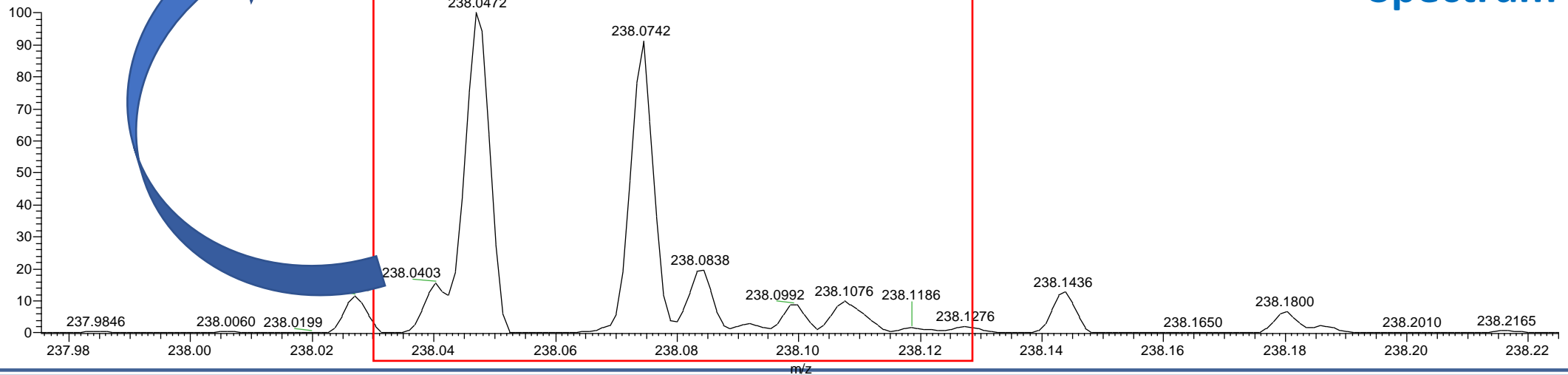
10/17/20 15:48:34

RT: 0.00 - 14.00



Cebolla_10ppb_AIF #7-6147 RT: 7.28 AV: 3071 NL: 1.39E5
T: FTMS + p ESI Full ms [100.0000-1000.0000]

Spectrum



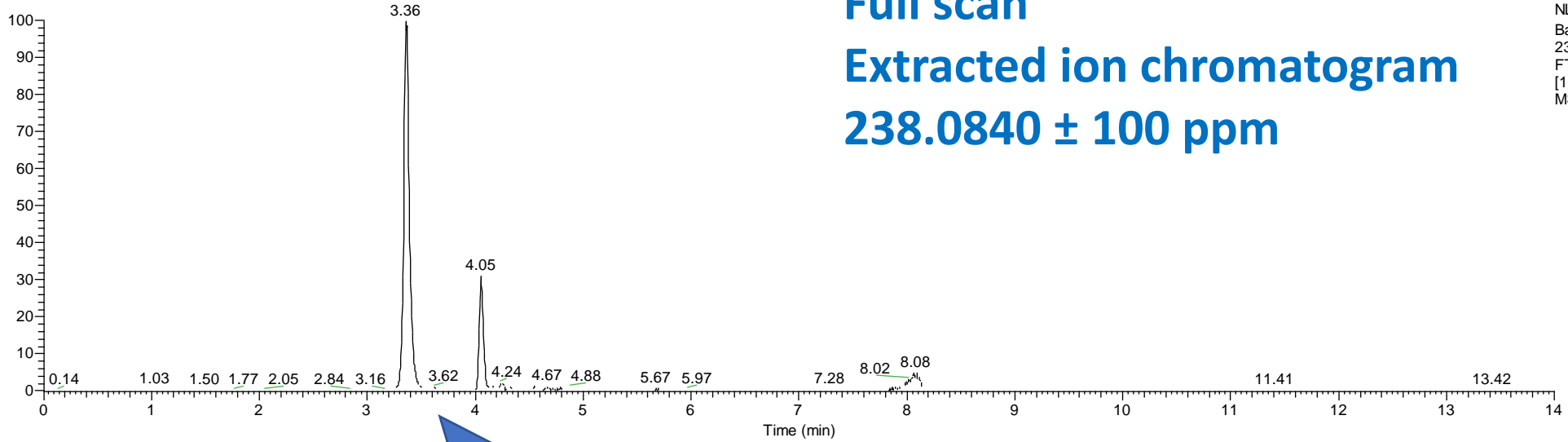
Onion

High-resolution mass spectrometry

D:\data\...\Cebolla_10ppb_AIF

10/17/20 15:48:34

RT: 0.00 - 14.00

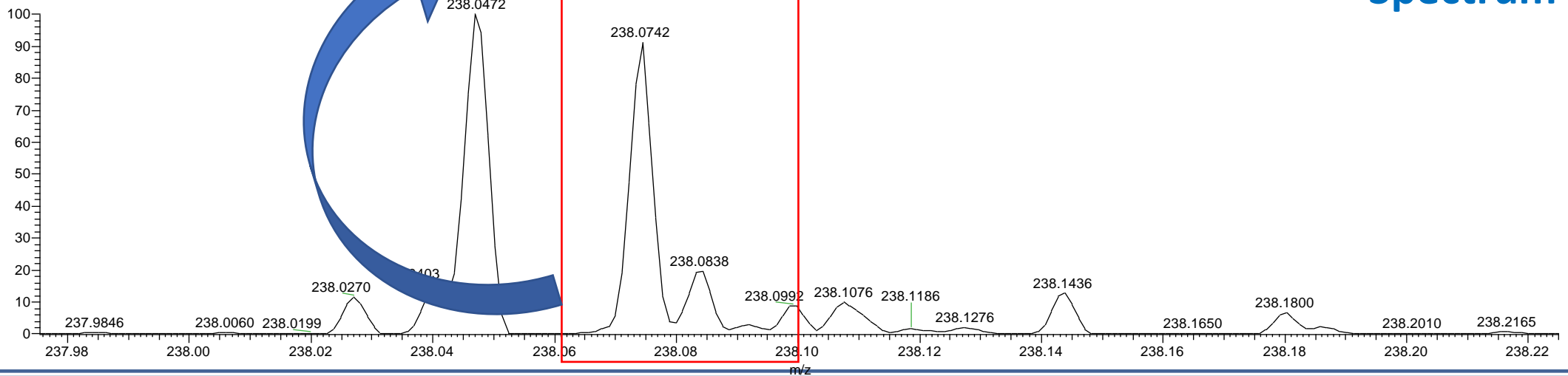


Full scan
Extracted ion chromatogram
238.0840 ± 100 ppm

NL: 2.86E7
Base Peak m/z=
238.06009-238.10771 F:
FTMS + p ESI Full ms
[100.0000-1000.0000]
MS Cebolla_10ppb_AIF

Cebolla_10ppb_AIF #7-6147 RT: 0.02-14.00 AV: 3071 NL: 1
T: FTMS + p ESI Full ms [100.0000-1000.0000]

Spectrum



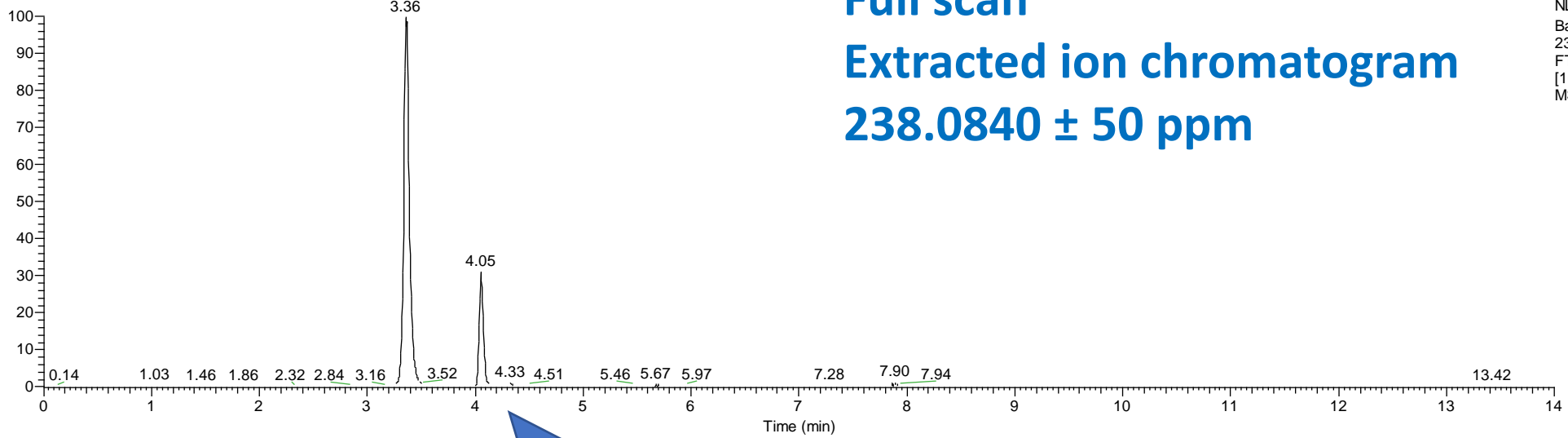
Onion

High-resolution mass spectrometry

D:\data...\Cebolla_10ppb_AIF

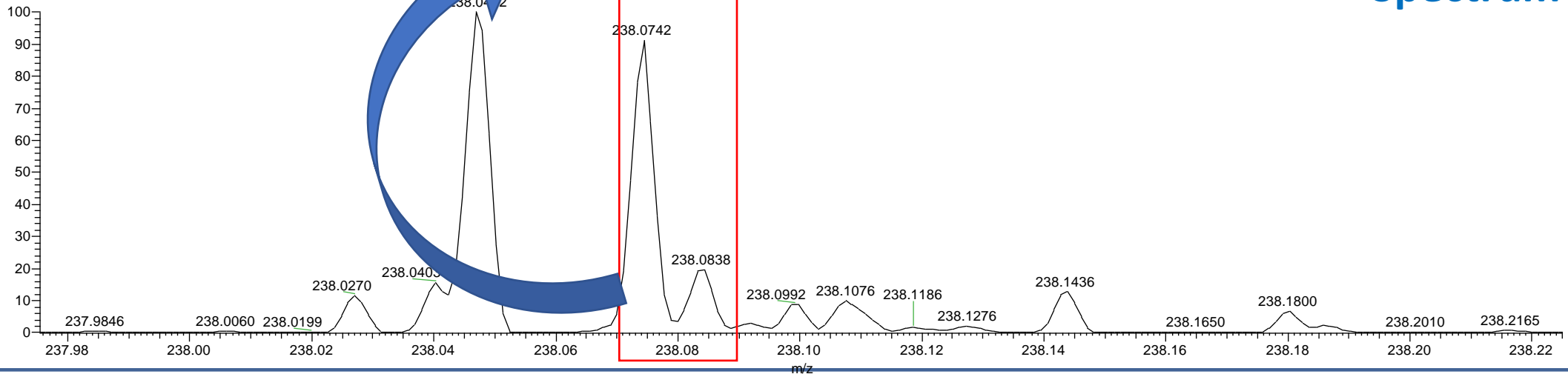
10/17/20 15:48:34

RT: 0.00 - 14.00



Cebolla_10ppb_AIF #7-6147 RT: 0.02-14.00 AV: 3071 NL: 1.39E5
T: FTMS + p ESI Full ms [100.0000-1000.0000]

Spectrum



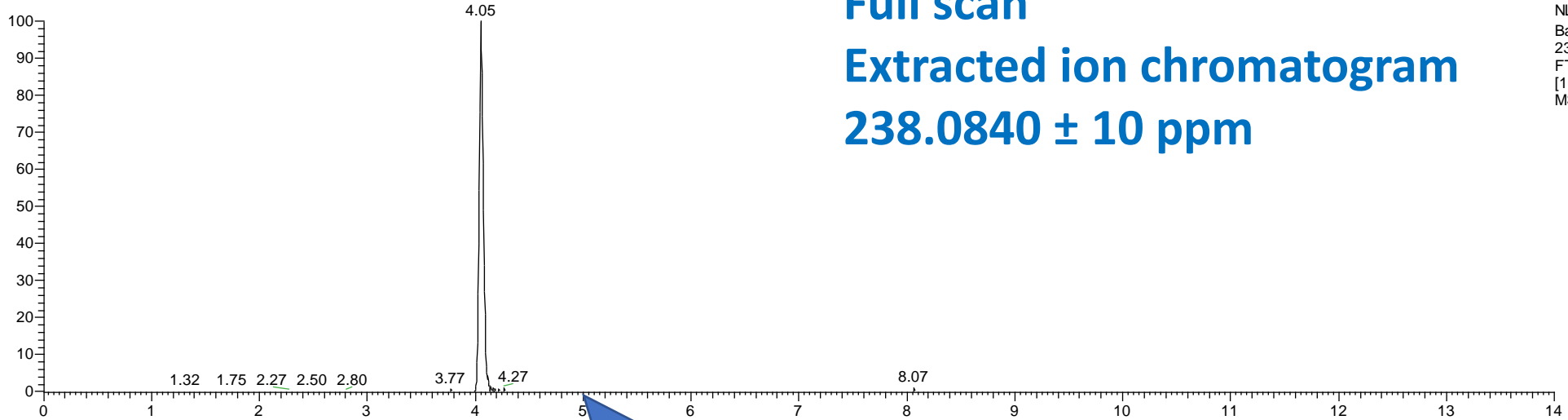
Onion

High-resolution mass spectrometry

D:\data...\Cebolla_10ppb_AIF

10/17/20 15:48:34

RT: 0.00 - 14.00

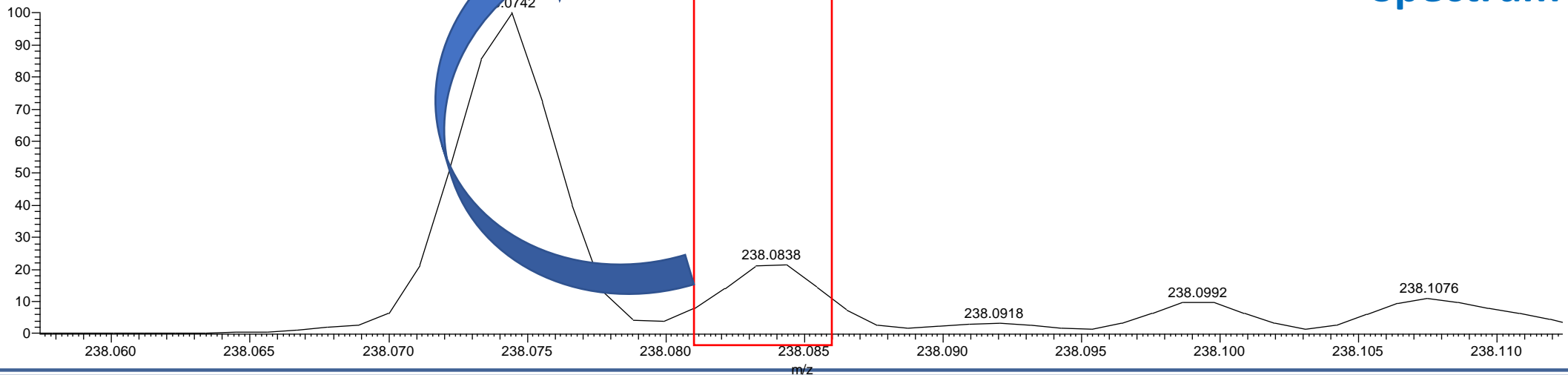


Full scan
Extracted ion chromatogram
238.0840 ± 10 ppm

NL: 8.88E6
Base Peak m/z=
238.08152-238.08628 F:
FTMS + p ESI Full ms
[100.0000-1000.0000]
MS Cebolla_10ppb_AIF

Cebolla_10ppb_AIF #7-6147 RT: 0.02-14.00 AV: 3071 NL: 1.27E5
T: FTMS + p ESI Full ms [100.0000-1000.0000]

Spectrum



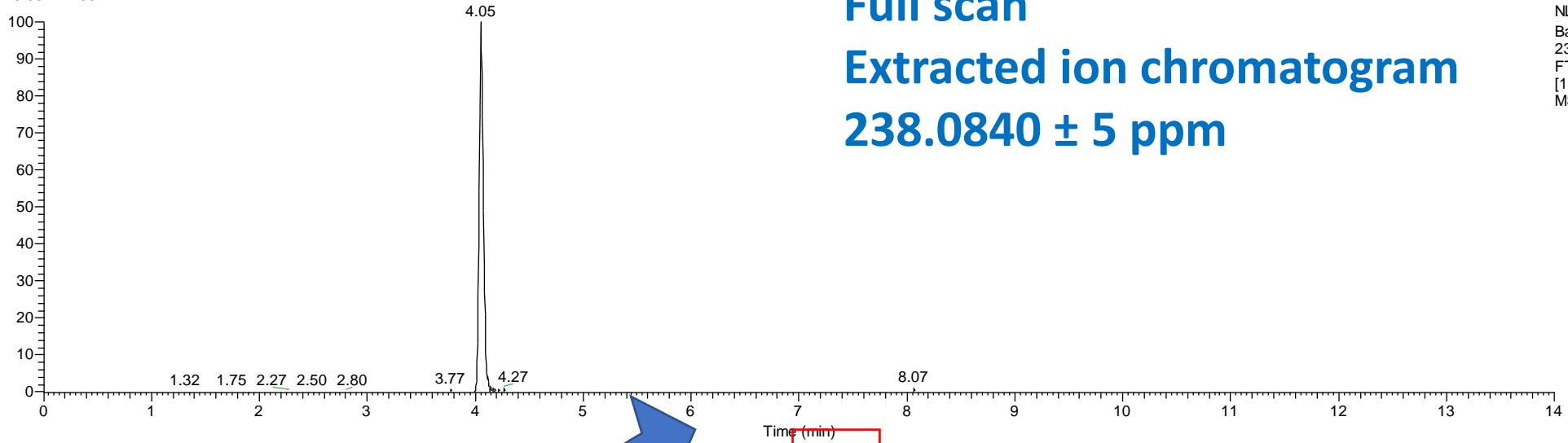
Onion

High-resolution mass spectrometry

D:\data\...\Cebolla_10ppb_AIF

10/17/20 15:48:34

RT: 0.00 - 14.00

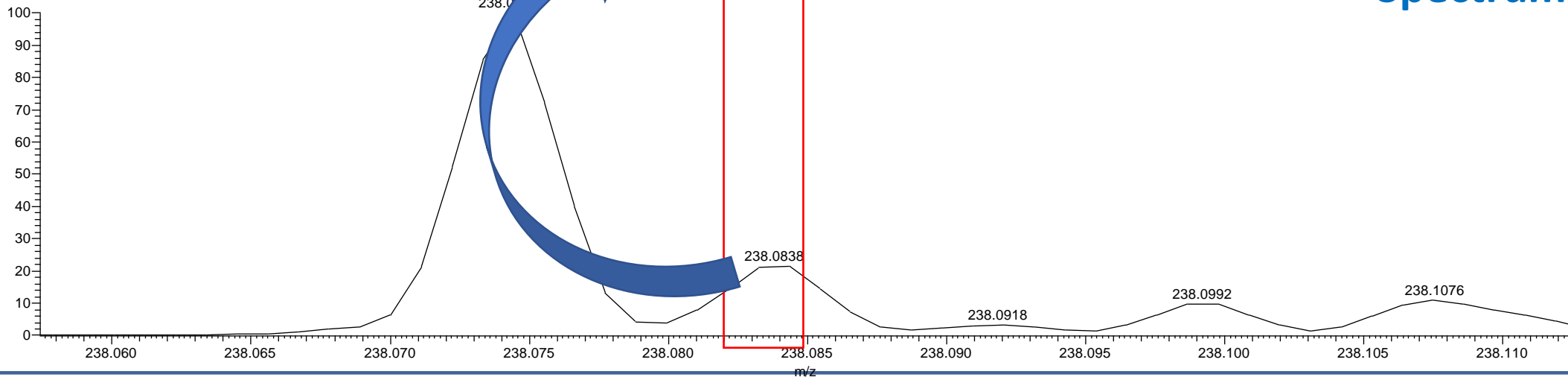


Full scan
Extracted ion chromatogram
238.0840 ± 5 ppm

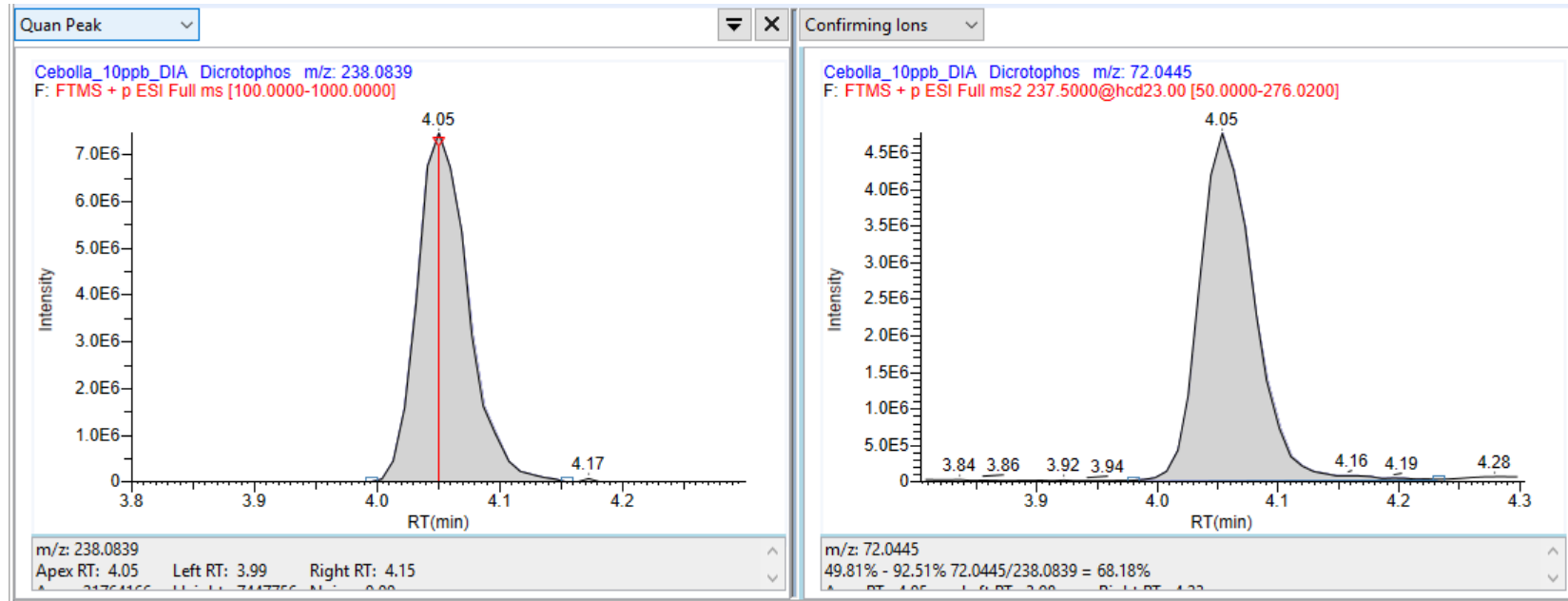
NL: 8.88E6
 Base Peak m/z=
 238.08271-238.08509 F:
 FTMS + p ESI Full ms
 [100.0000-1000.0000]
 MS Cebolla_10ppb_AIF

Cebolla_10ppb_AIF #7-6147 RT: 0.02-14.00 AV: 3071 NL: 1.27E5
 T: FTMS + p ESI Full ms [100.0000-1000.0000]

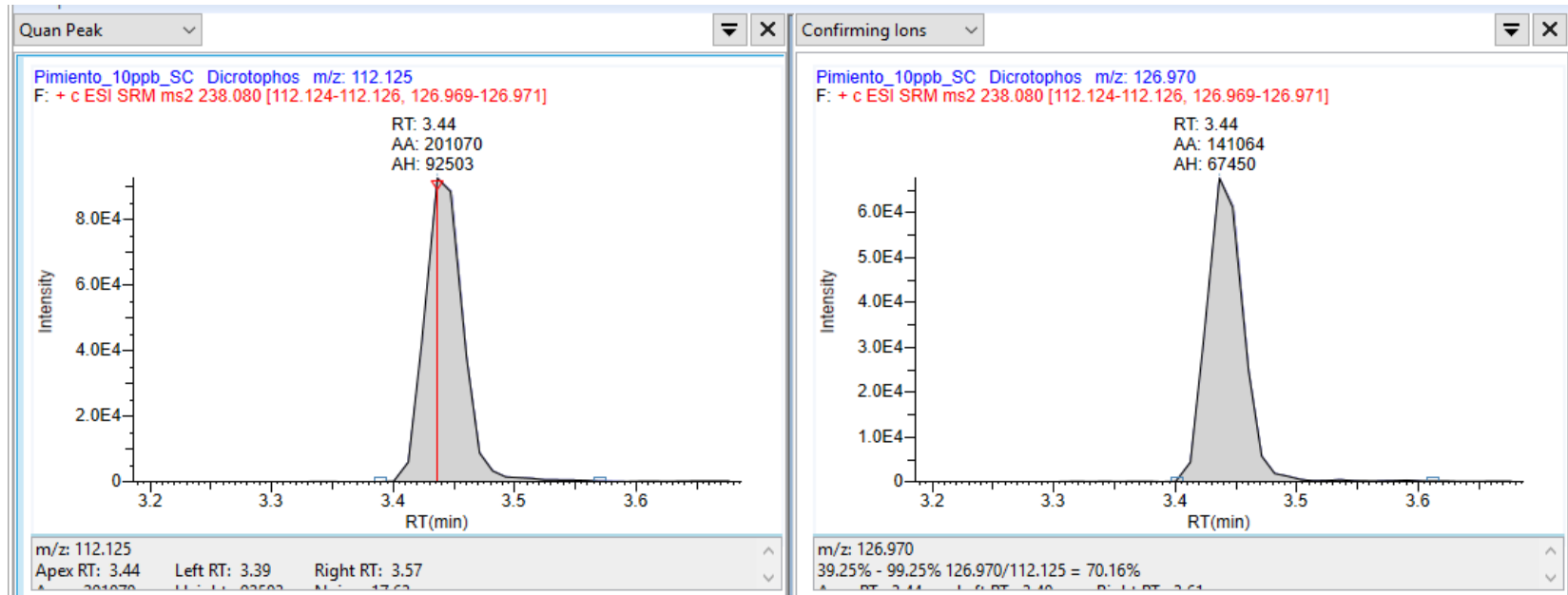
Spectrum



Dicrotophos In HRMS →

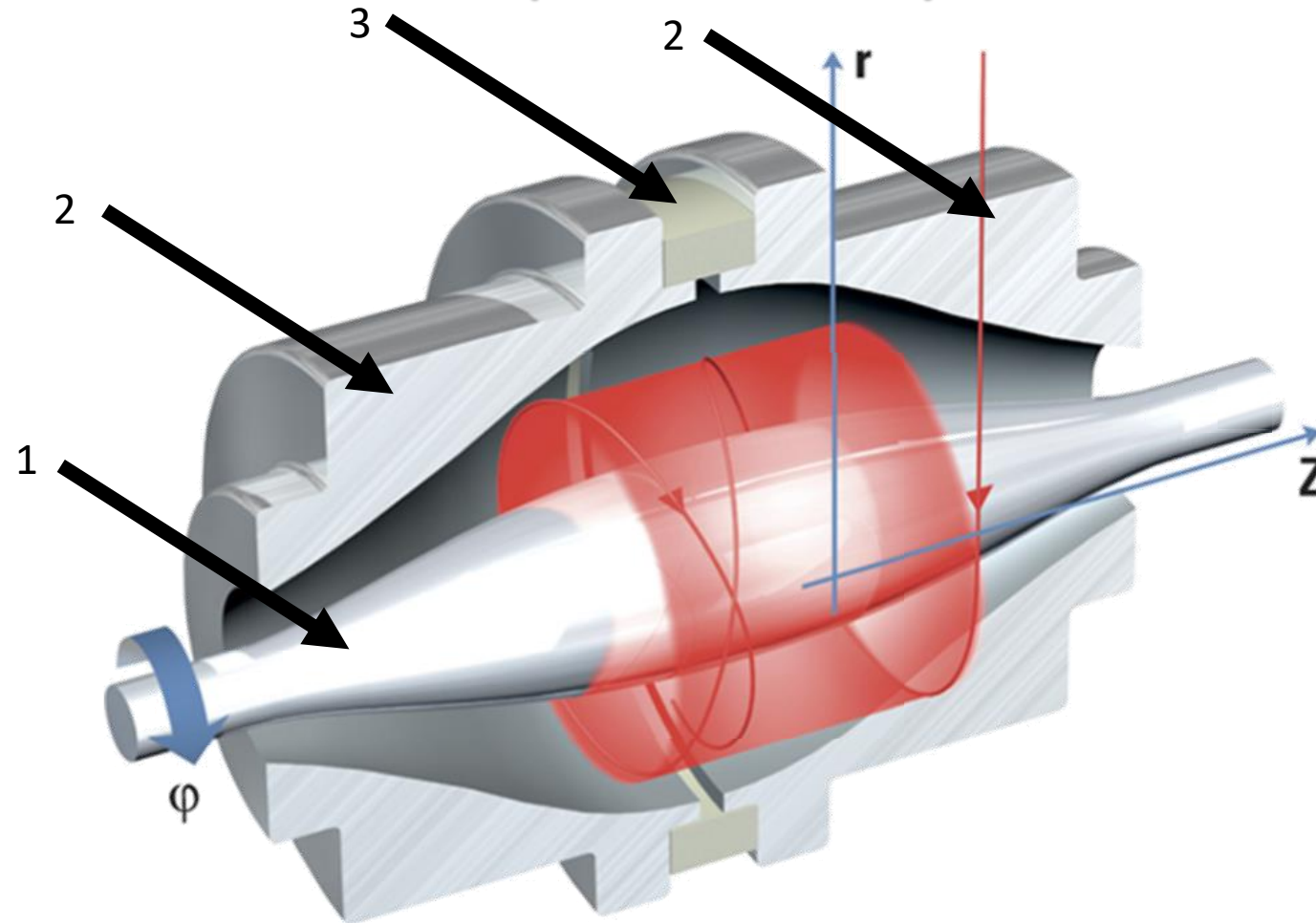


Dicrotophos In QQQ →



High-resolution mass spectrometry

Orbitrap mass analyser



1- central electrode

2- half of outer electrode

3- isolating ring

High-resolution mass spectrometry

Orbitrap mass analyser

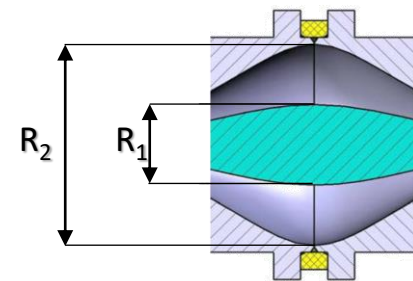


$$\omega = \sqrt{\frac{e}{m/z} \cdot k}$$



$$m/z = \frac{e \cdot k}{\omega^2}$$

$$k = \sqrt{\frac{2 \cdot U_r}{R_m^2 \ln\left(\frac{R_2}{R_1}\right) - \frac{1}{2} [R_2^2 - R_1^2]}}$$



High-resolution mass spectrometry

Orbitrap mass analyser

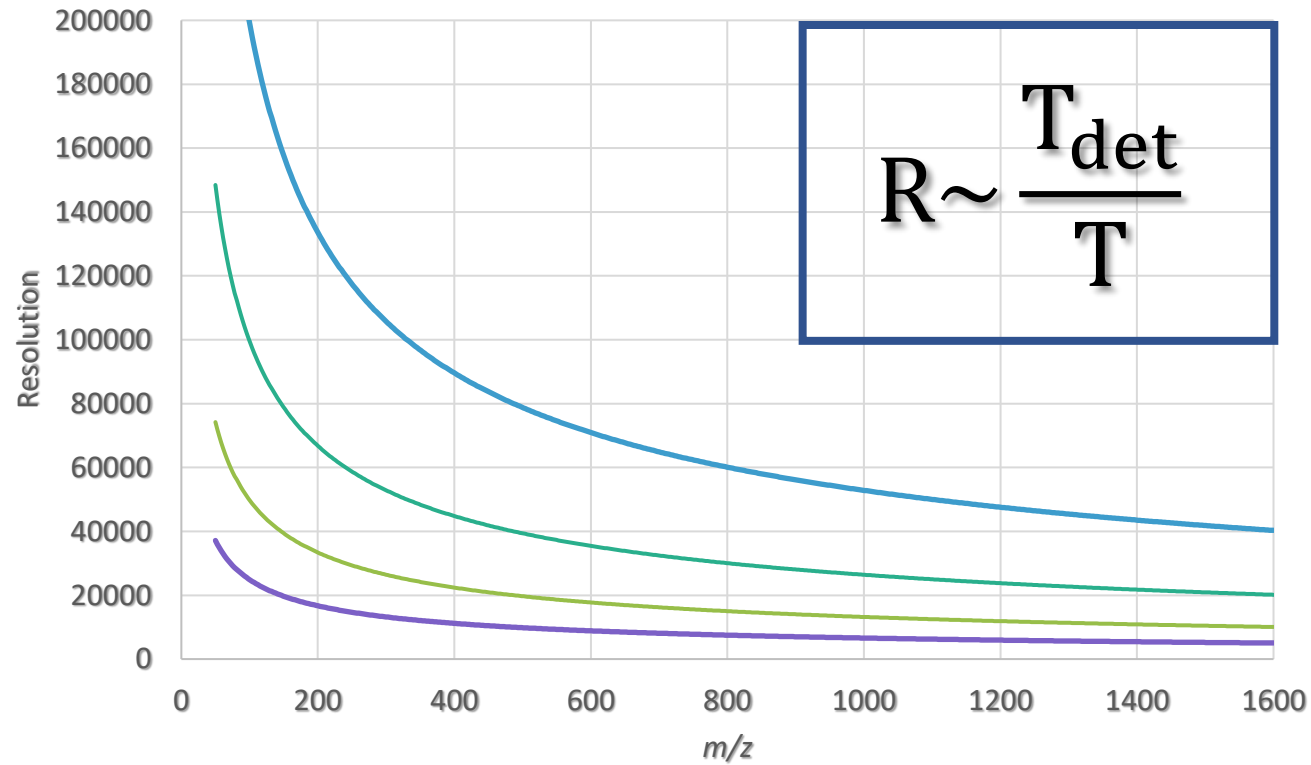


$$R \sim \frac{T_{\text{det}}}{T}$$

$$\omega = \sqrt{\frac{e}{m/z} \cdot k} \quad \Rightarrow \quad T = \frac{1}{\omega} \quad \Rightarrow \quad m/z = e \cdot k \cdot T^2 \quad \Rightarrow \quad T = \sqrt{\frac{m/z}{e \cdot k}}$$

High-resolution mass spectrometry

Orbitrap mass analyser



■ 660 ms (140 000 at m/z 200)

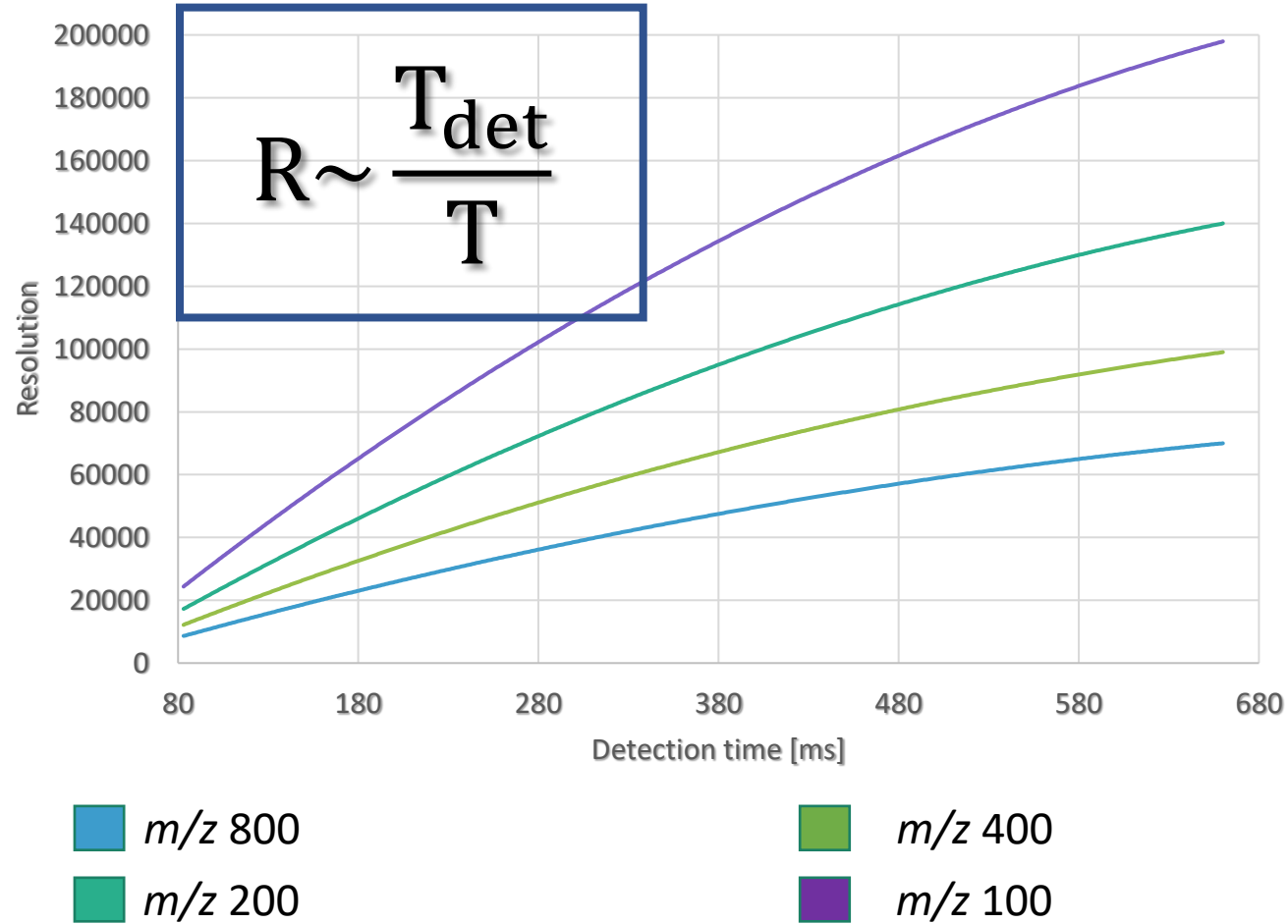
■ 143 ms (35 000 at m/z 200)

■ 270 ms (70 000 at m/z 200)

■ 83 ms (17 500 at m/z 200)

High-resolution mass spectrometry

Orbitrap mass analyser



High-resolution mass spectrometry

Orbitrap mass analyser

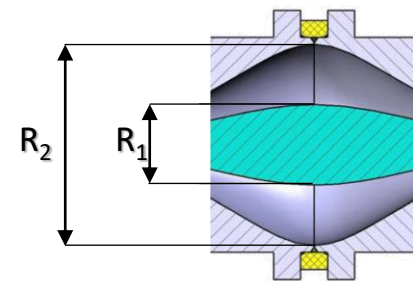


$$\omega = \sqrt{\frac{e}{m/z} \cdot k}$$



$$m/z = \frac{e \cdot k}{\omega^2}$$

$$k = \sqrt{\frac{2 \cdot U_r}{R_m^2 \ln\left(\frac{R_2}{R_1}\right) - \frac{1}{2} [R_2^2 - R_1^2]}}$$



High-resolution mass spectrometry



Orbitrap Exploris 240



High-resolution mass spectrometry

QExactive/QExactive Focus		Exploris 240/ Exploris 120	
Mass resolution at m/z 200	Scan rate	Mass resolution at m/z 200	Scan rate
17 500	12 Hz	15 000	22 Hz
35 000	7 Hz	30 000	12 Hz
		45 000*	10 Hz
70 000	3 Hz	60 000	7 Hz
140 000*	1.5 Hz	120 000	3 Hz
		240 000**	1.5 Hz

* Not available in QExactive Focus

** Not available in Exploris 120

Comparison of cycle times

Full scan MS @70K + AIF @ 70K (QExactive)



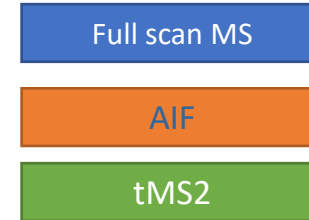
Full scan MS @60K + AIF @ 60K Orbitrap (Exploris 240 MS)



Full scan MS @70K + 10 tMS2 @ 17.5K (QExactive)



Full scan MS @60K + 10 tMS2 @ 15K (Orbitrap Exploris 240 MS)

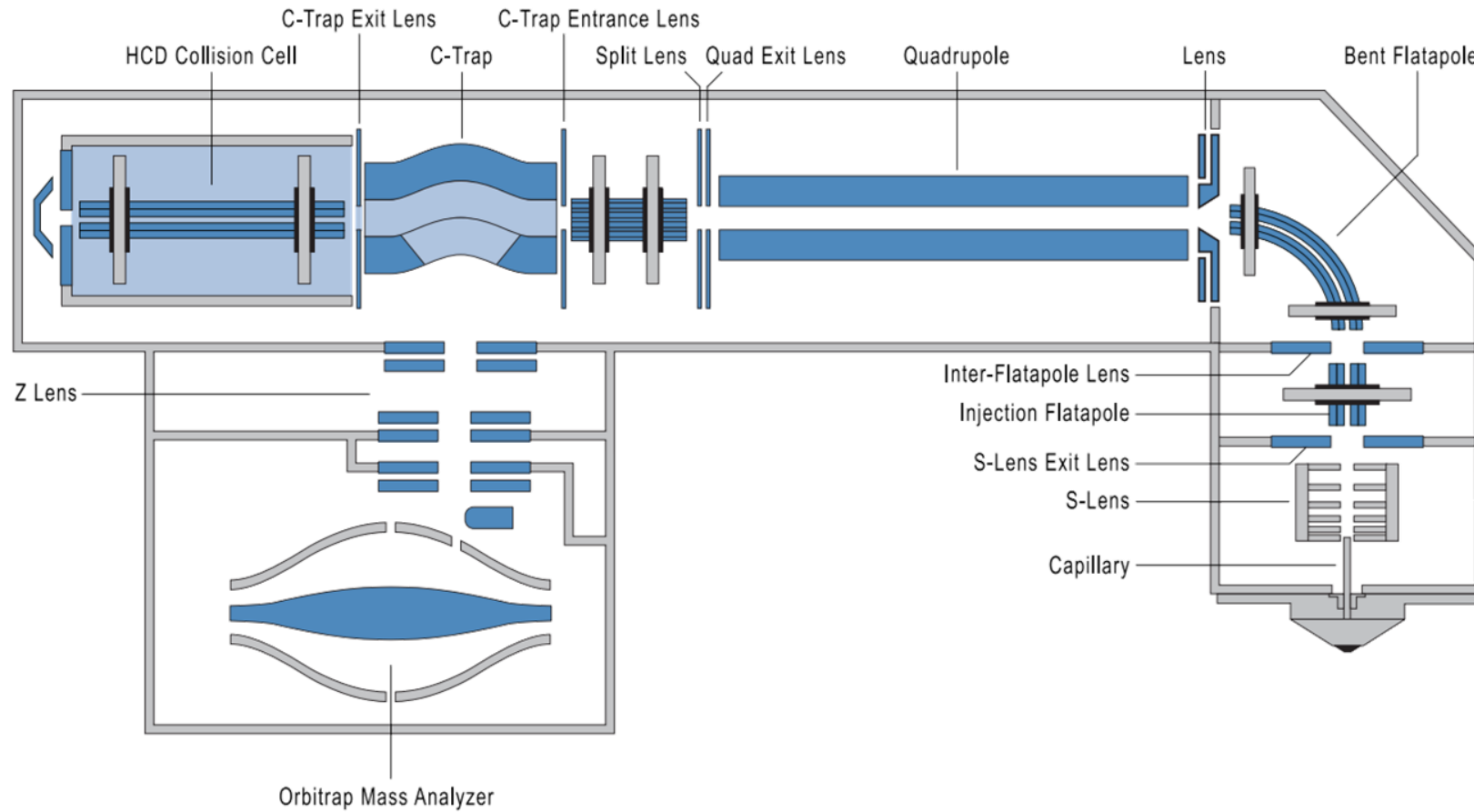


Orbitrap Exploris 240 MS

Vs

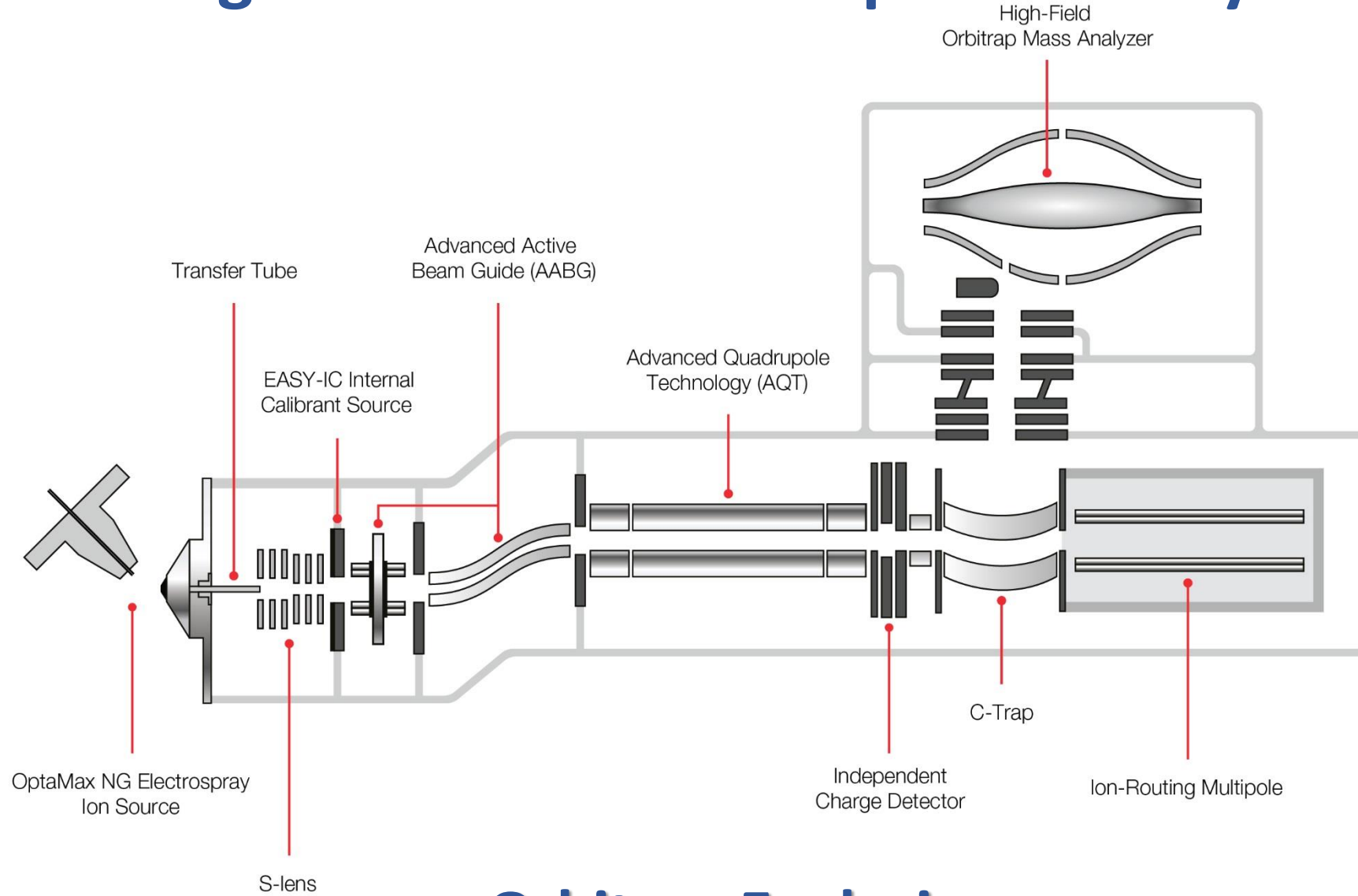
Thermo Scientific™ Q Exactive™ Hybrid Quadrupole-Orbitrap™ MS systems (Classic & Plus)

High-resolution mass spectrometry



Q Exactive

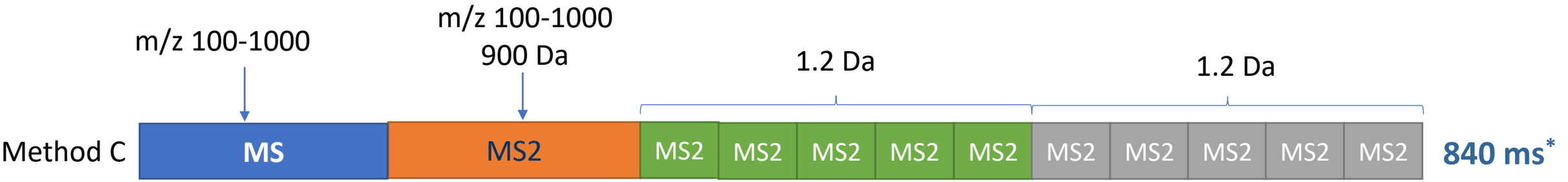
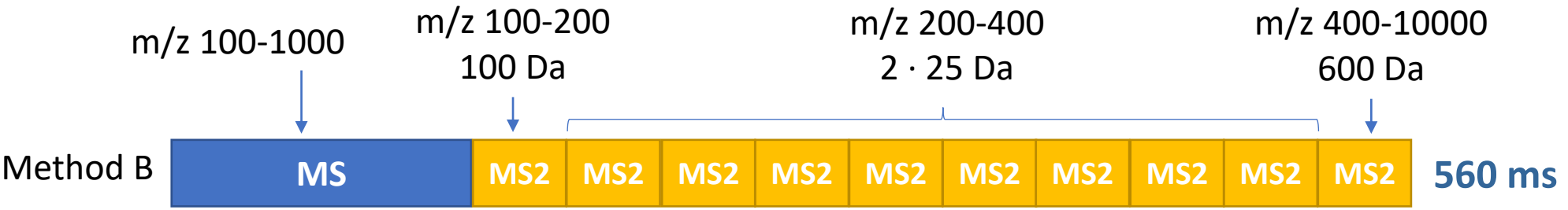
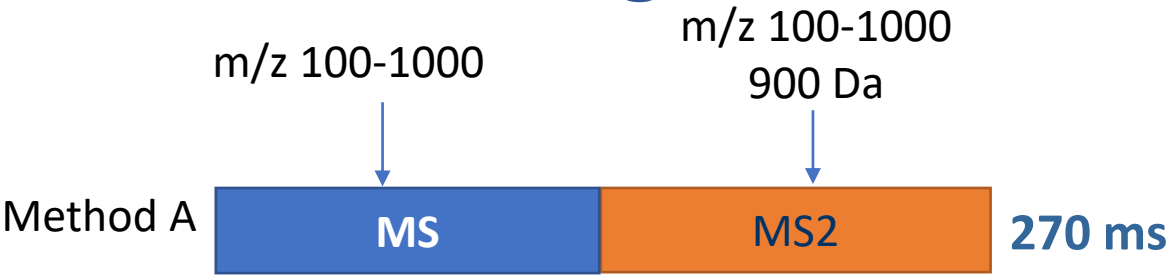
High-resolution mass spectrometry



Orbitrap Exploris

High-resolution mass spectrometry

- Full scan MS @ 60k
- AIF @ 60k
- DIA @ 15k
- tMS2 @ 15k
- ddMS2 @ 15k

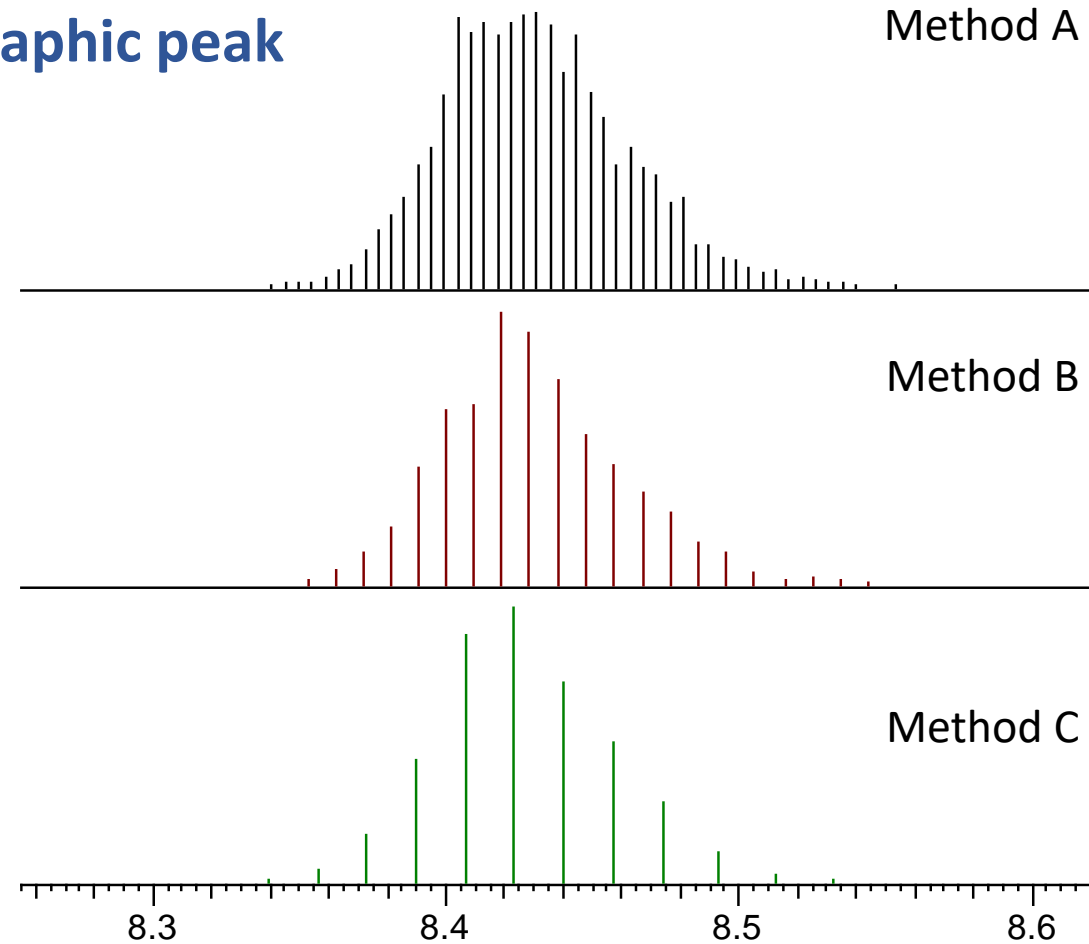


Target analysis 244 compounds
 Screening: ~900 compounds

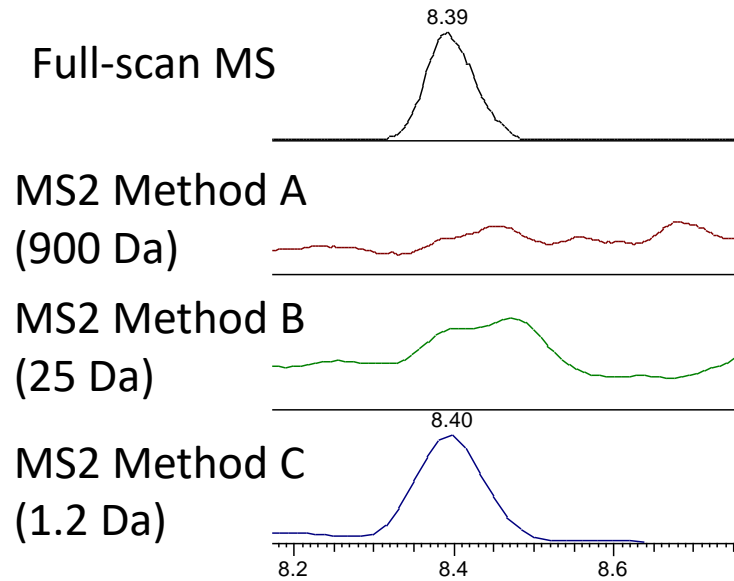
*average value

High-resolution mass spectrometry

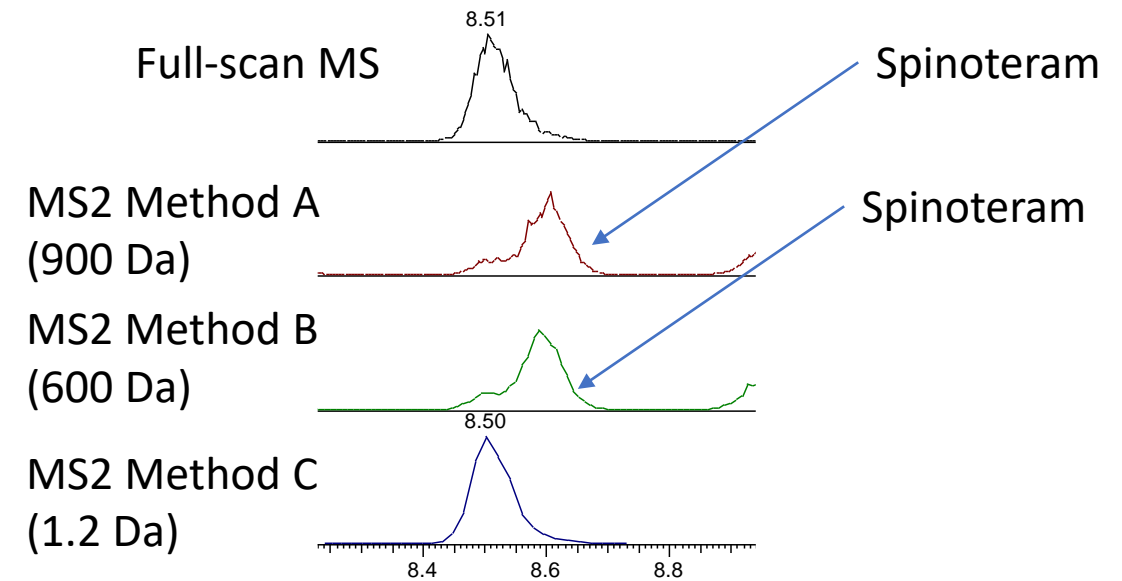
Data points per chromatographic peak



High-resolution mass spectrometry

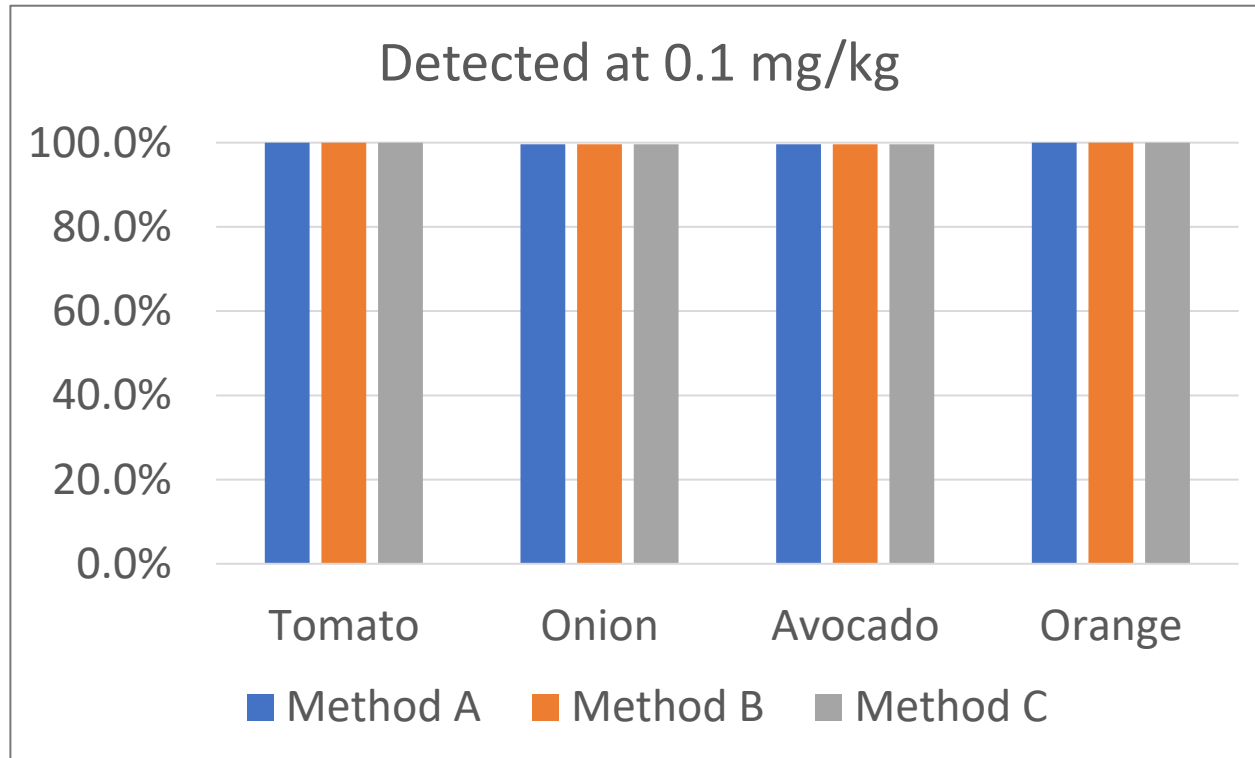
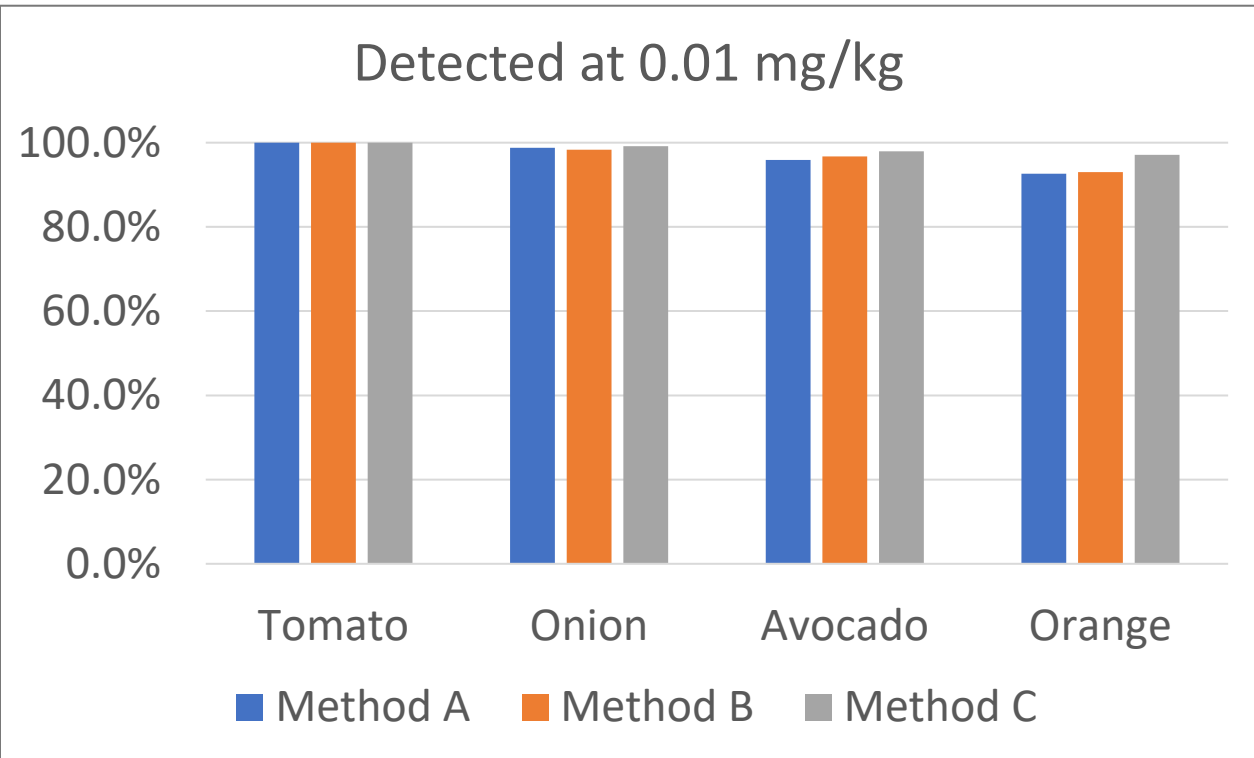


Dodine 0.01 mg/kg in orange
 MS m/z 228.2434 \pm 5ppm
 MS2 m/z 57.0700 \pm 5ppm

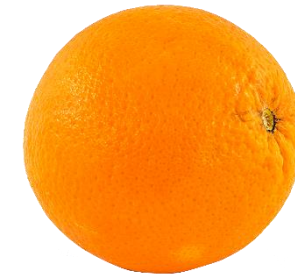


Spinosyn D 0.50 mg/kg in pure solvent
 MS m/z 746.4838 \pm 5ppm
 MS2 m/z 142.1226 \pm 5ppm

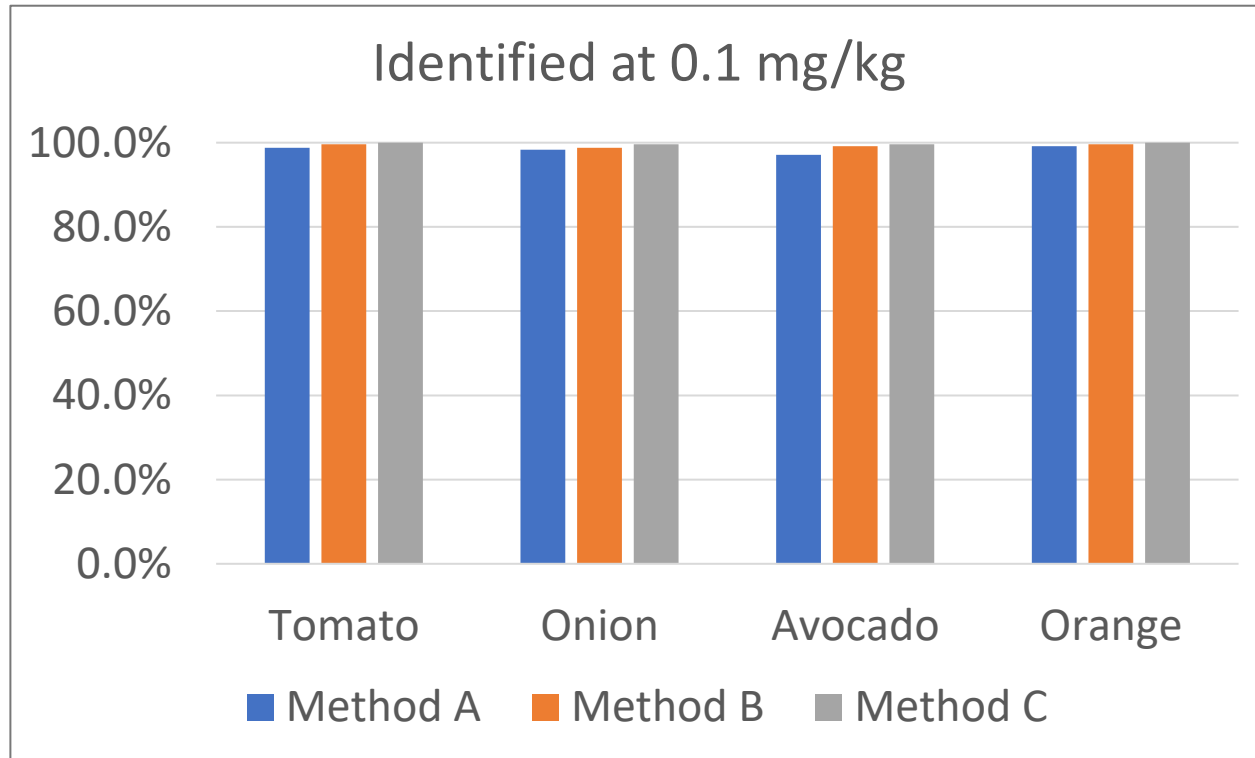
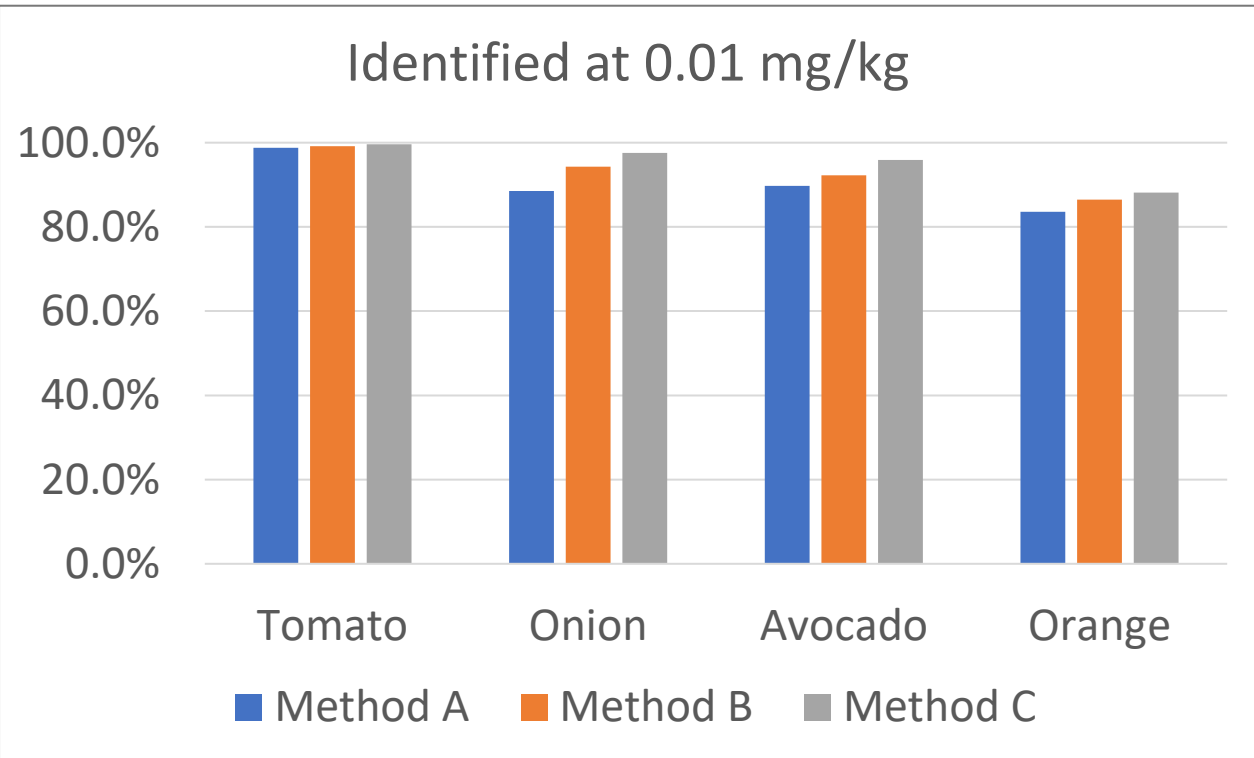
Target pesticides- validation



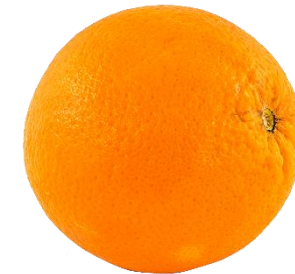
244 pesticides



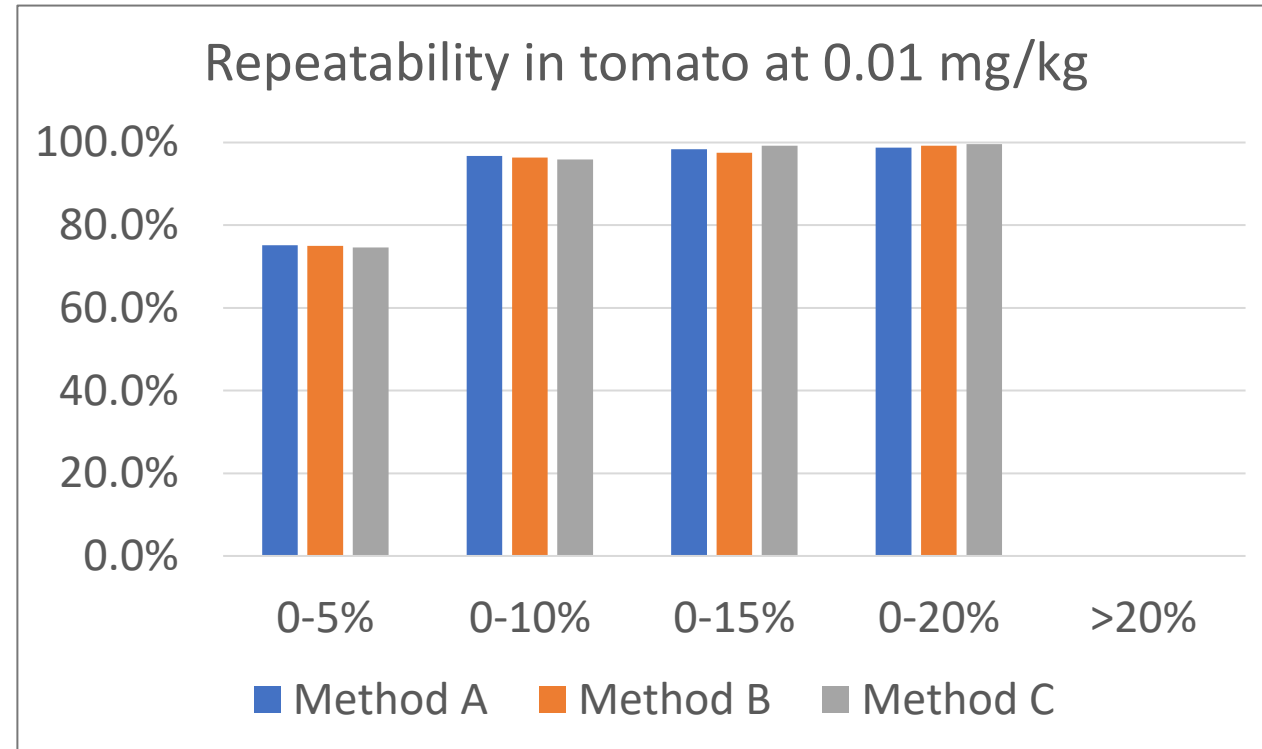
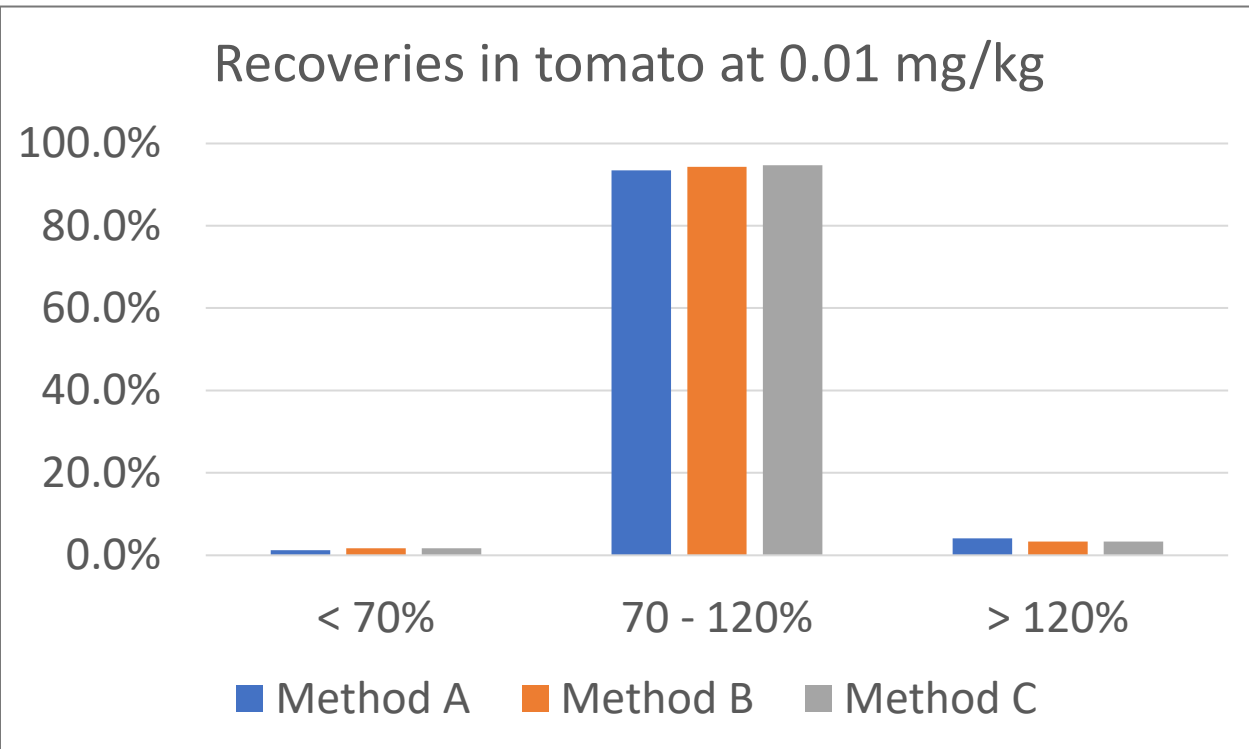
Target pesticides- validation



244 pesticides



Target pesticides- validation

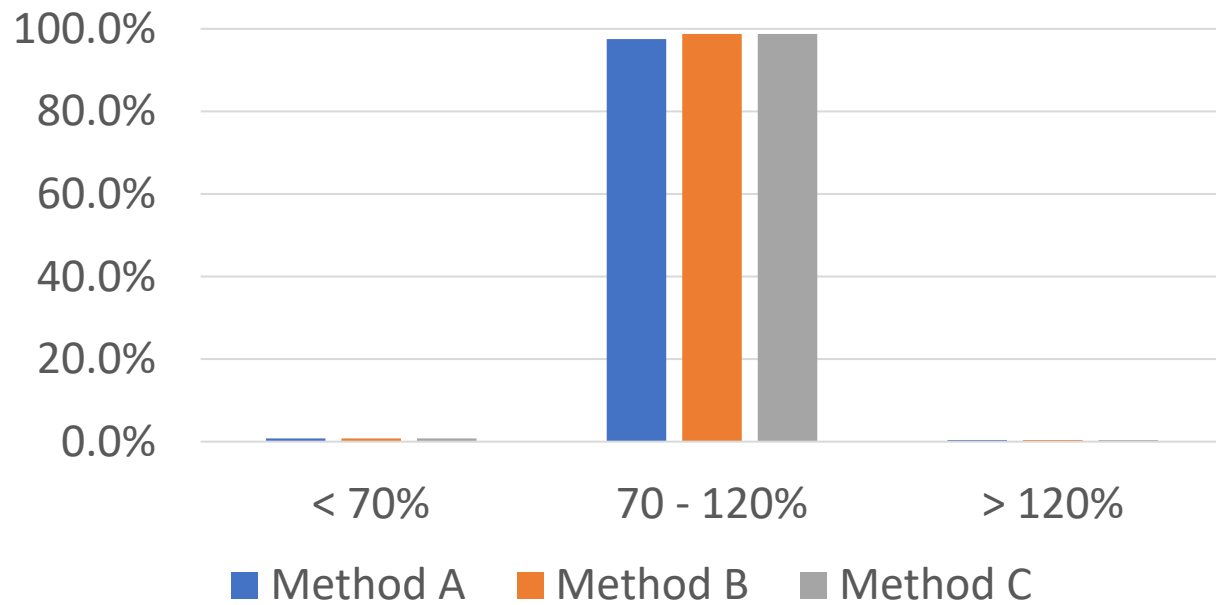


244 pesticides

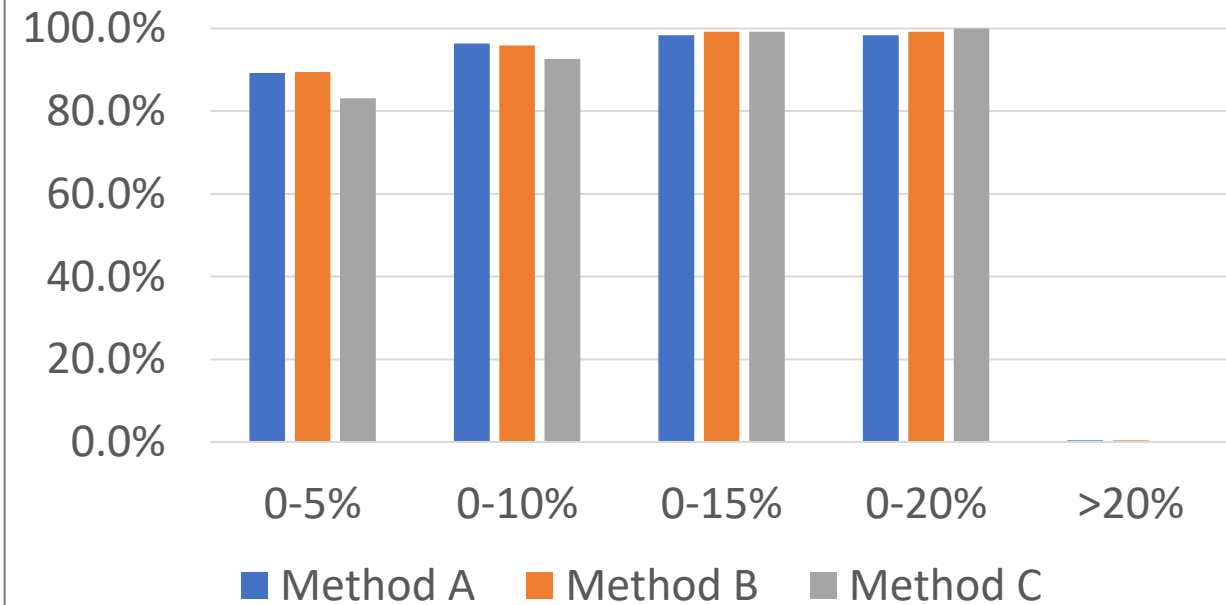


Target pesticides- validation

Recoveries in tomato at 0.1 mg/kg



Repeatability in tomato at 0.1 mg/kg

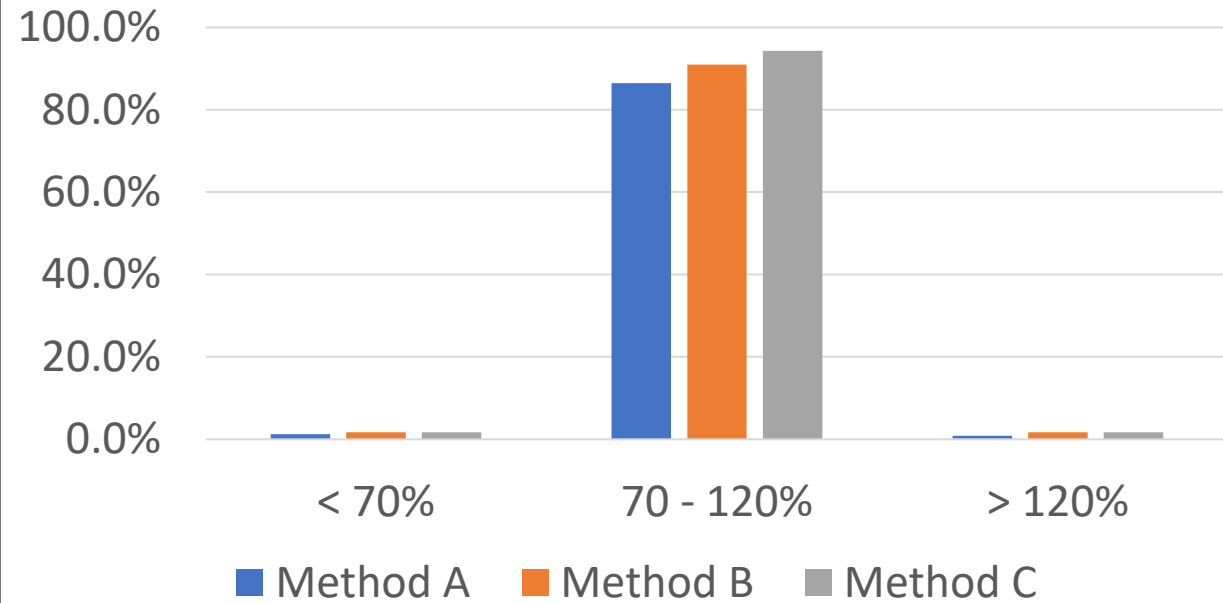


244 pesticides

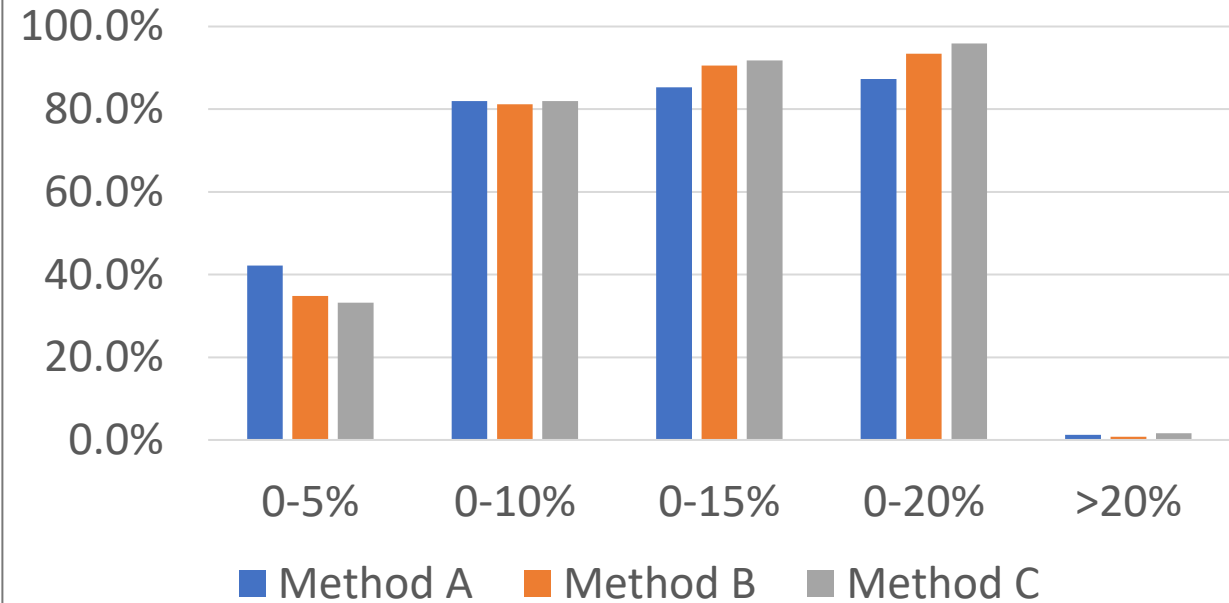


Target pesticides- validation

Recoveries in onion at 0.01 mg/kg

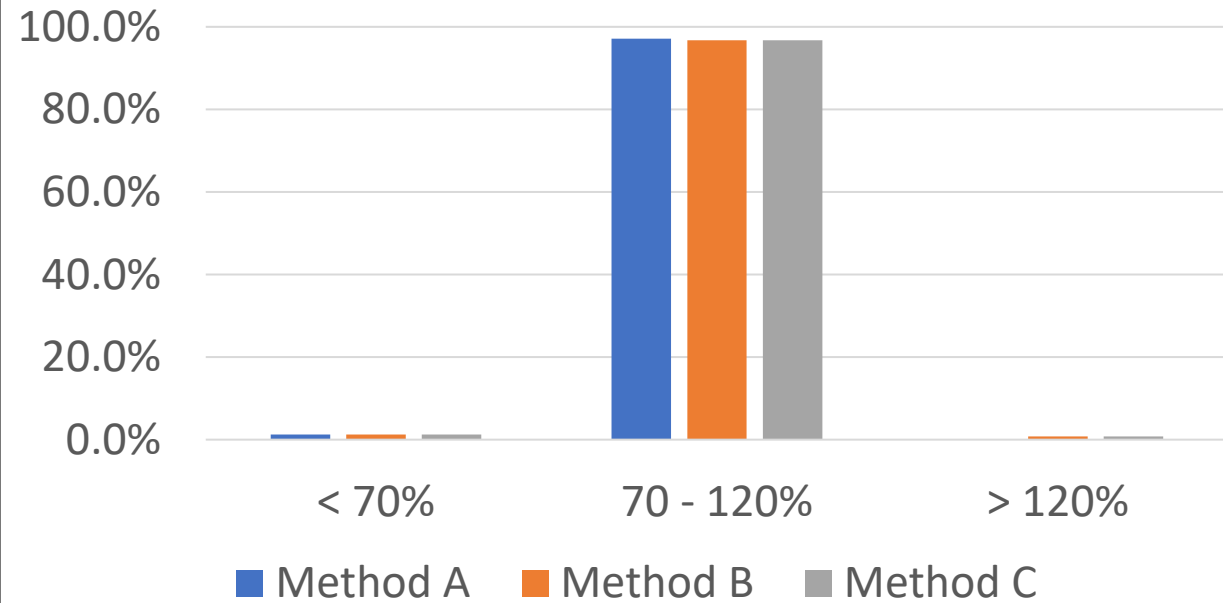


Repeatability in onion at 0.01 mg/kg

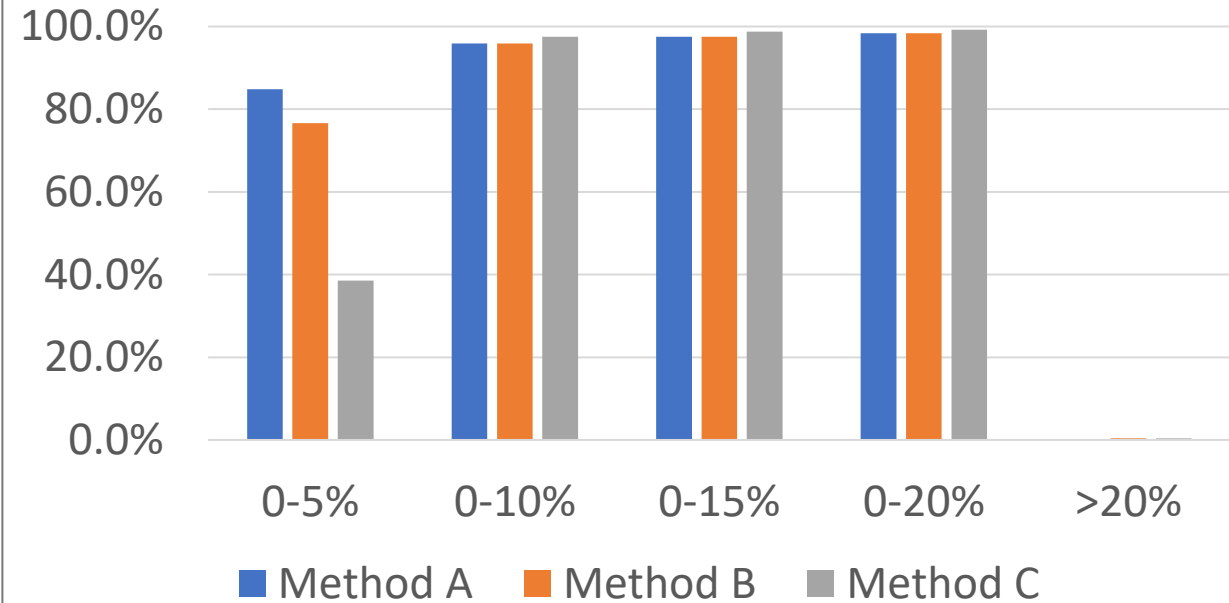


Target pesticides- validation

Recoveries in onion at 0.1 mg/kg



Repeatability in onion at 0.1 mg/kg

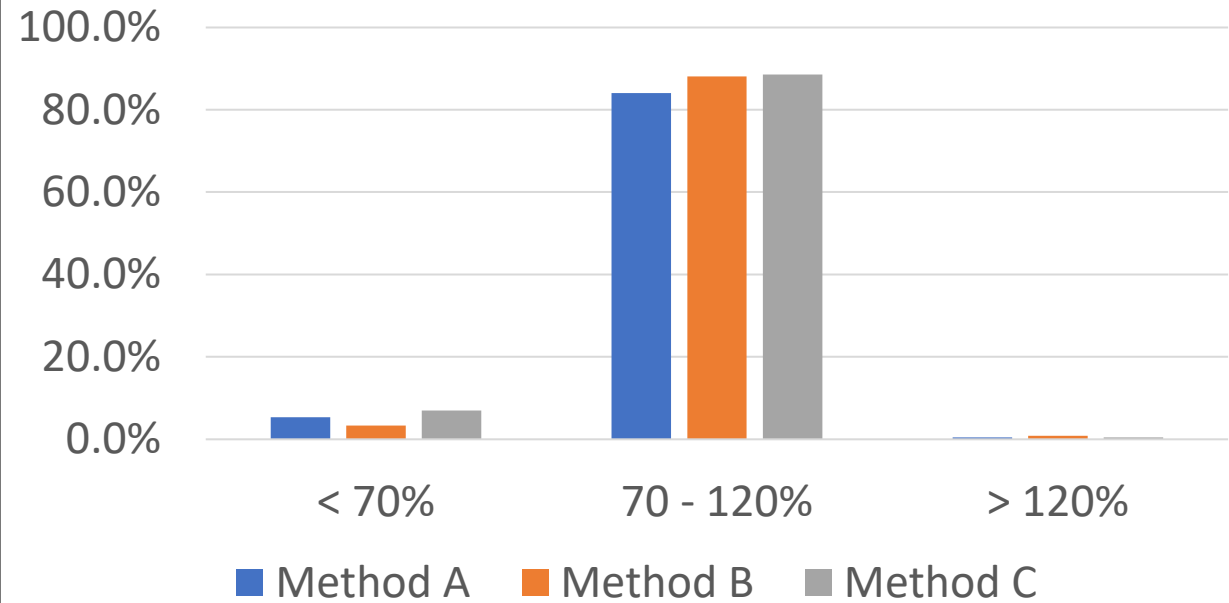


244 pesticides

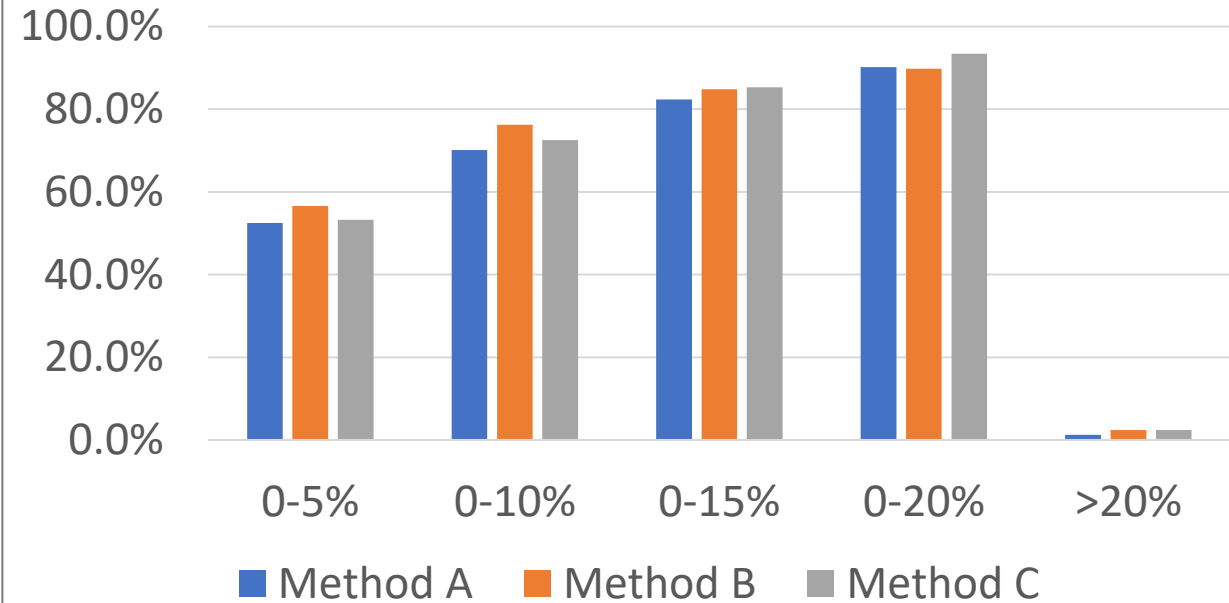


Target pesticides- validation

Recoveries in avocado at 0.01 mg/kg



Repeatability in avocado at 0.01 mg/kg

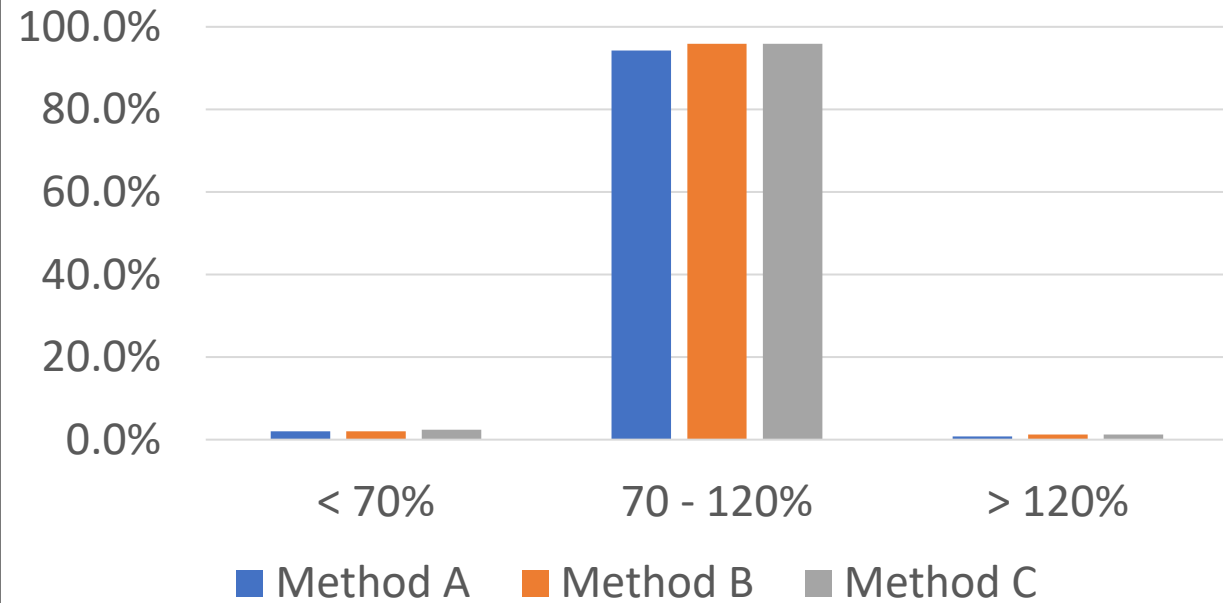


244 pesticides

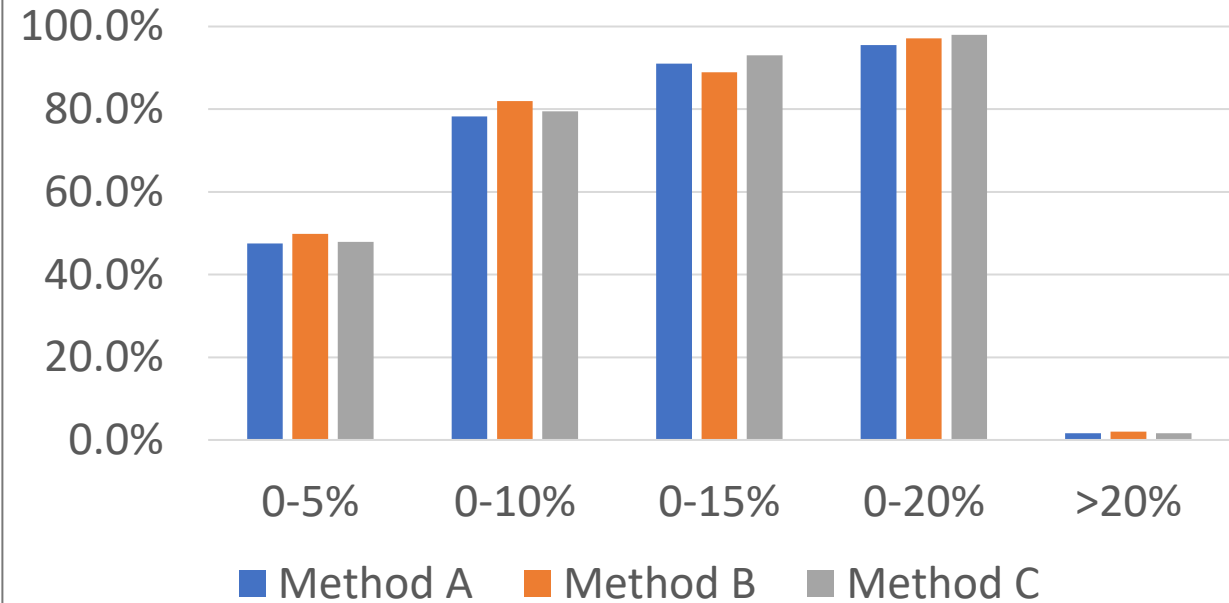


Target pesticides- validation

Recoveries in avocado at 0.1 mg/kg



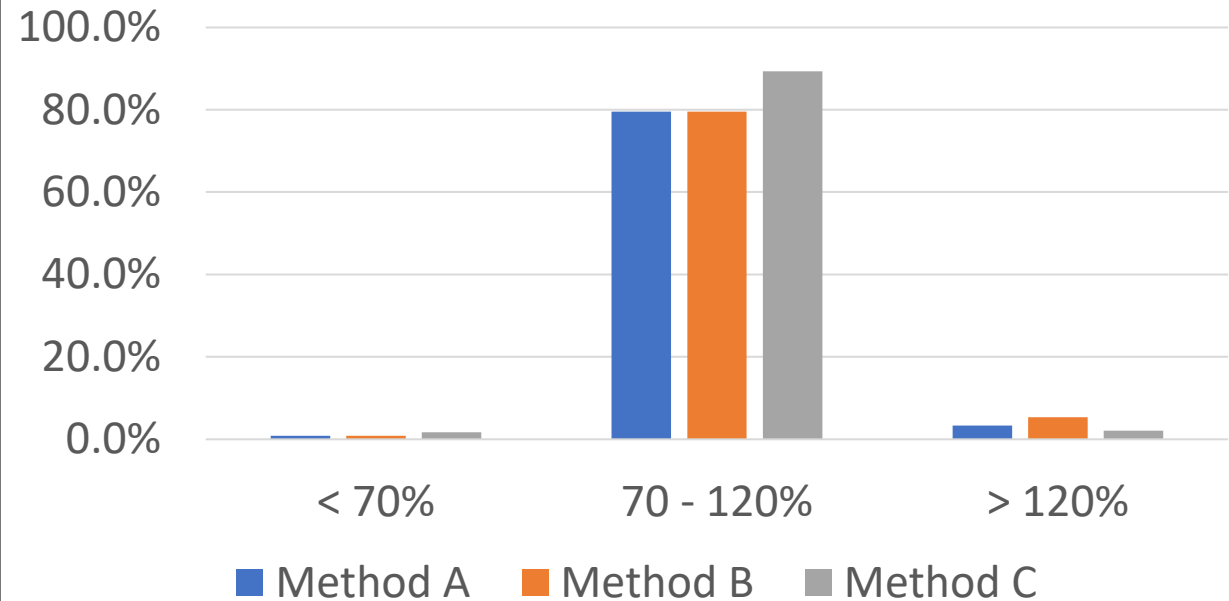
Repeatability in avocado at 0.1 mg/kg



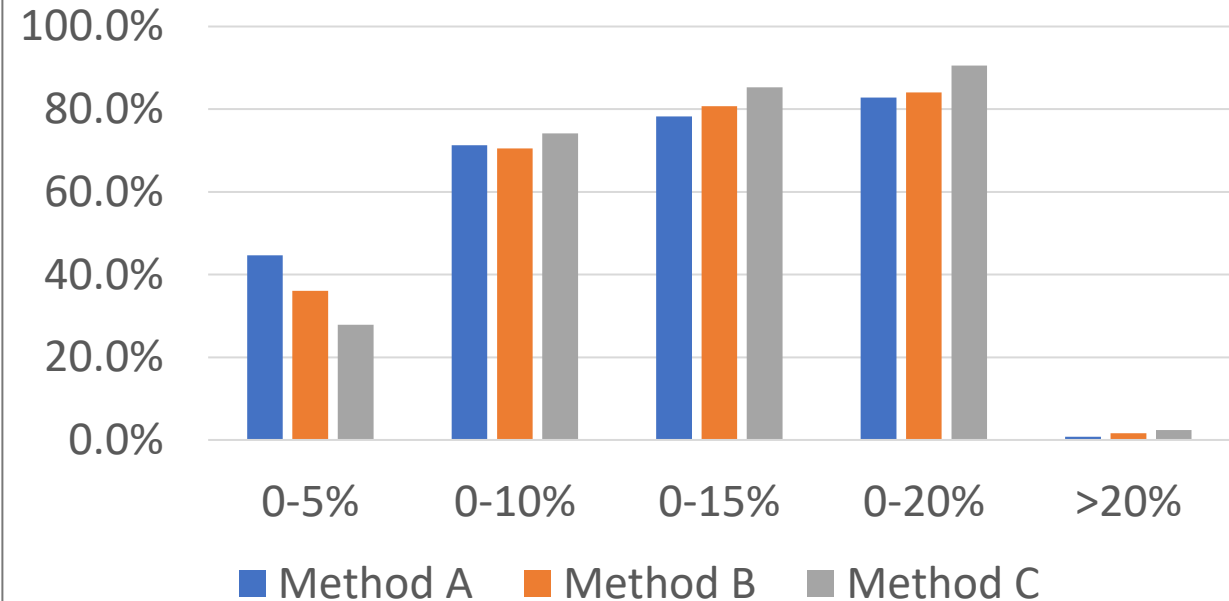
244 pesticides

Target pesticides- validation

Recoveries in orange at 0.01 mg/kg



Repeatability in orange at 0.01 mg/kg

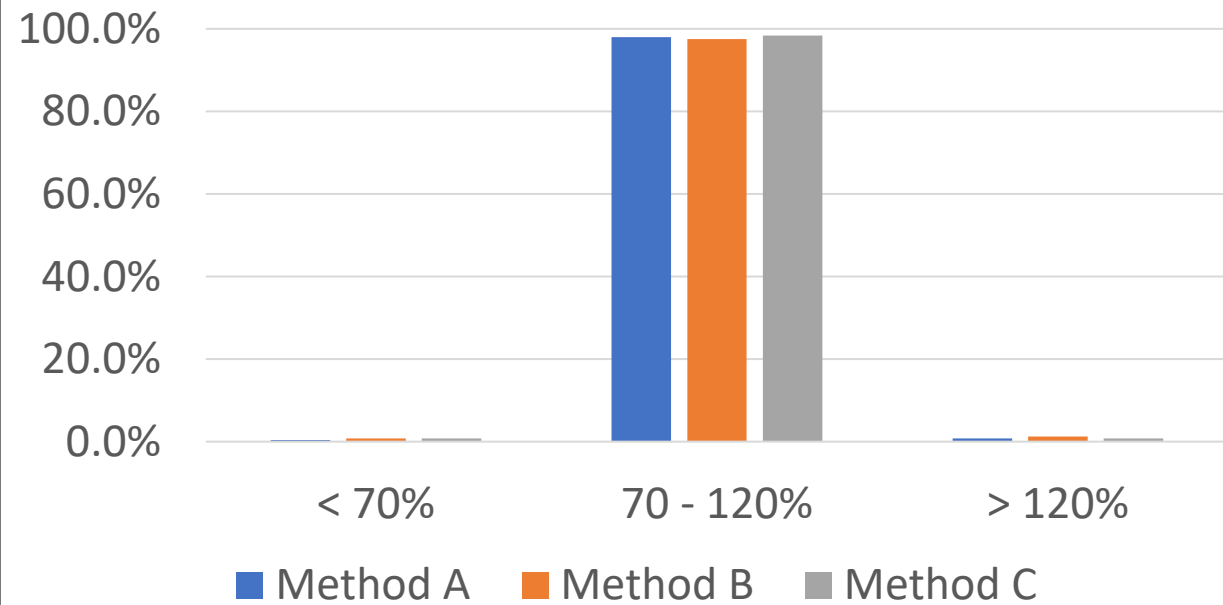


244 pesticides

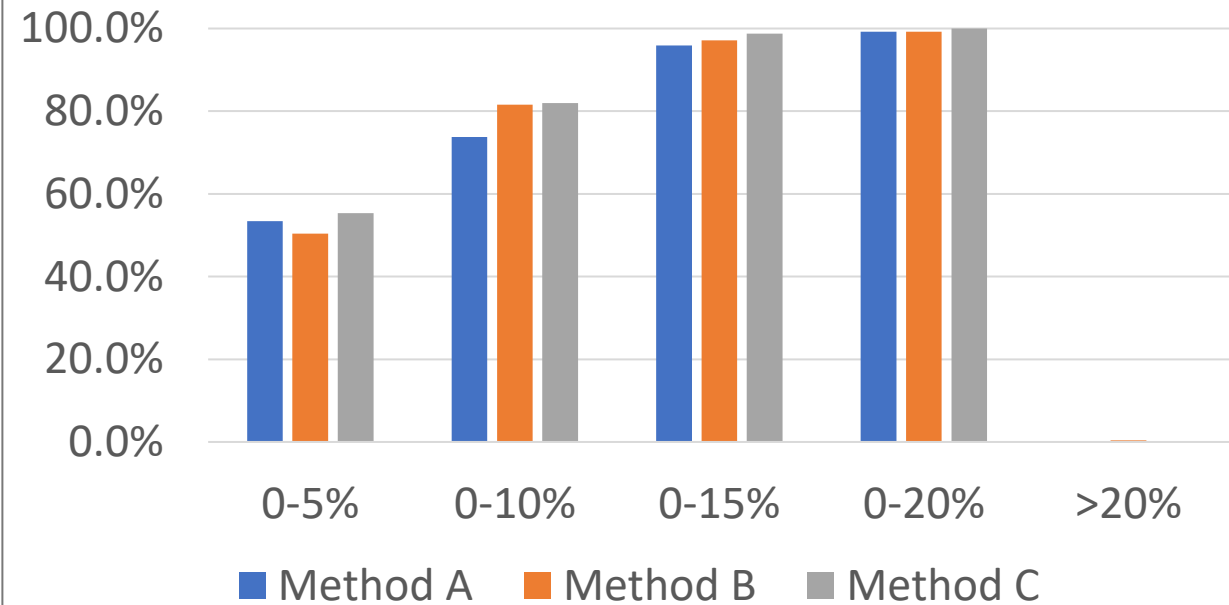


Target pesticides- validation

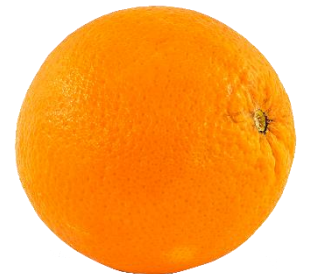
Recoveries in orange at 0.1 mg/kg



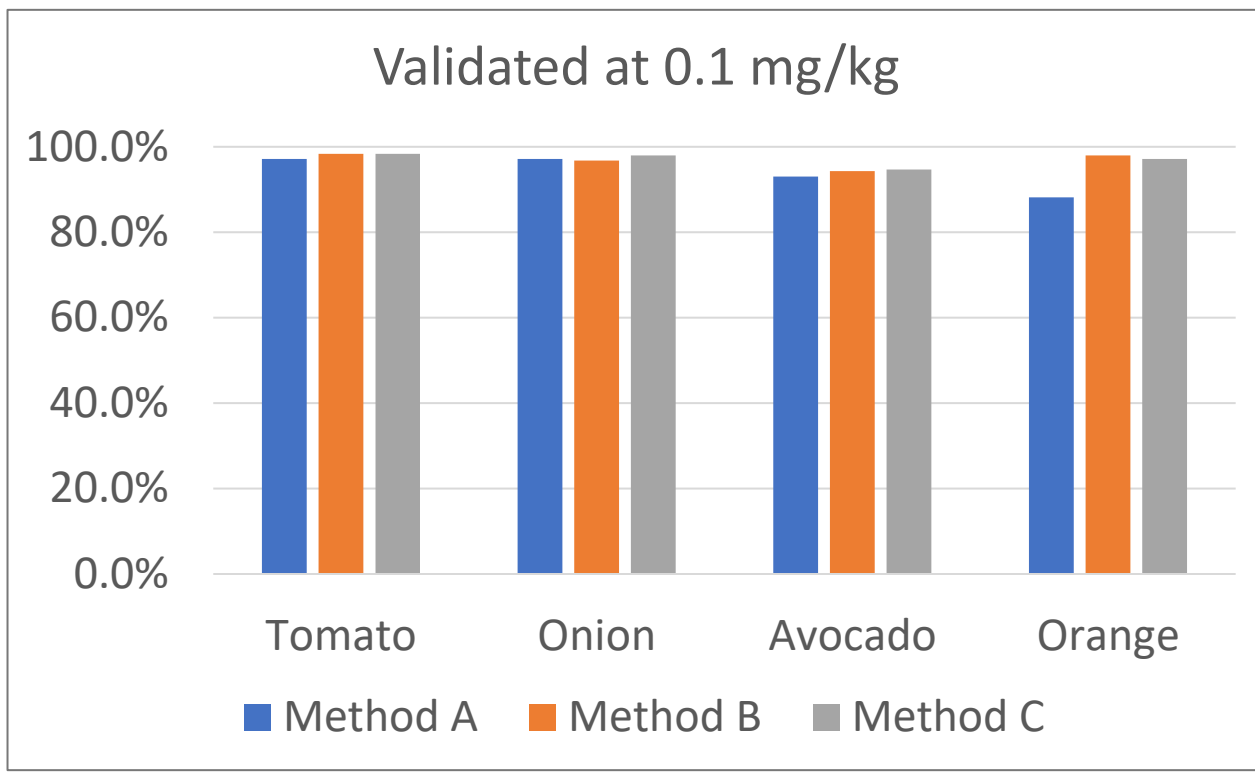
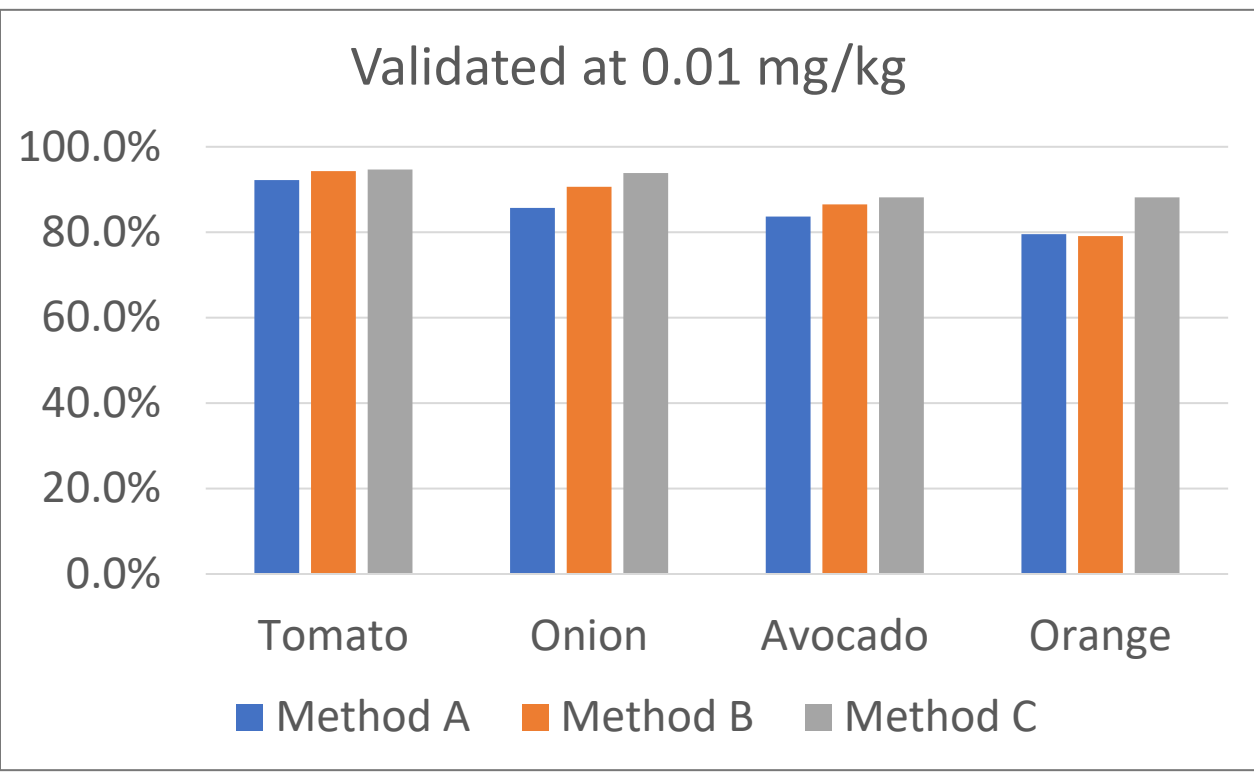
Repeatability in orange at 0.1 mg/kg



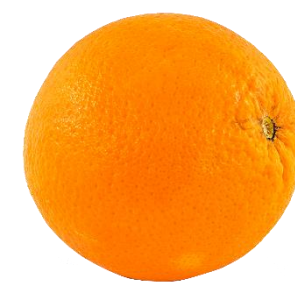
244 pesticides



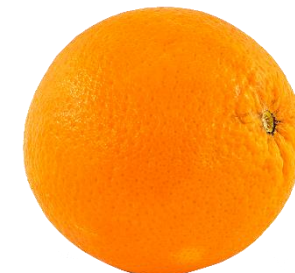
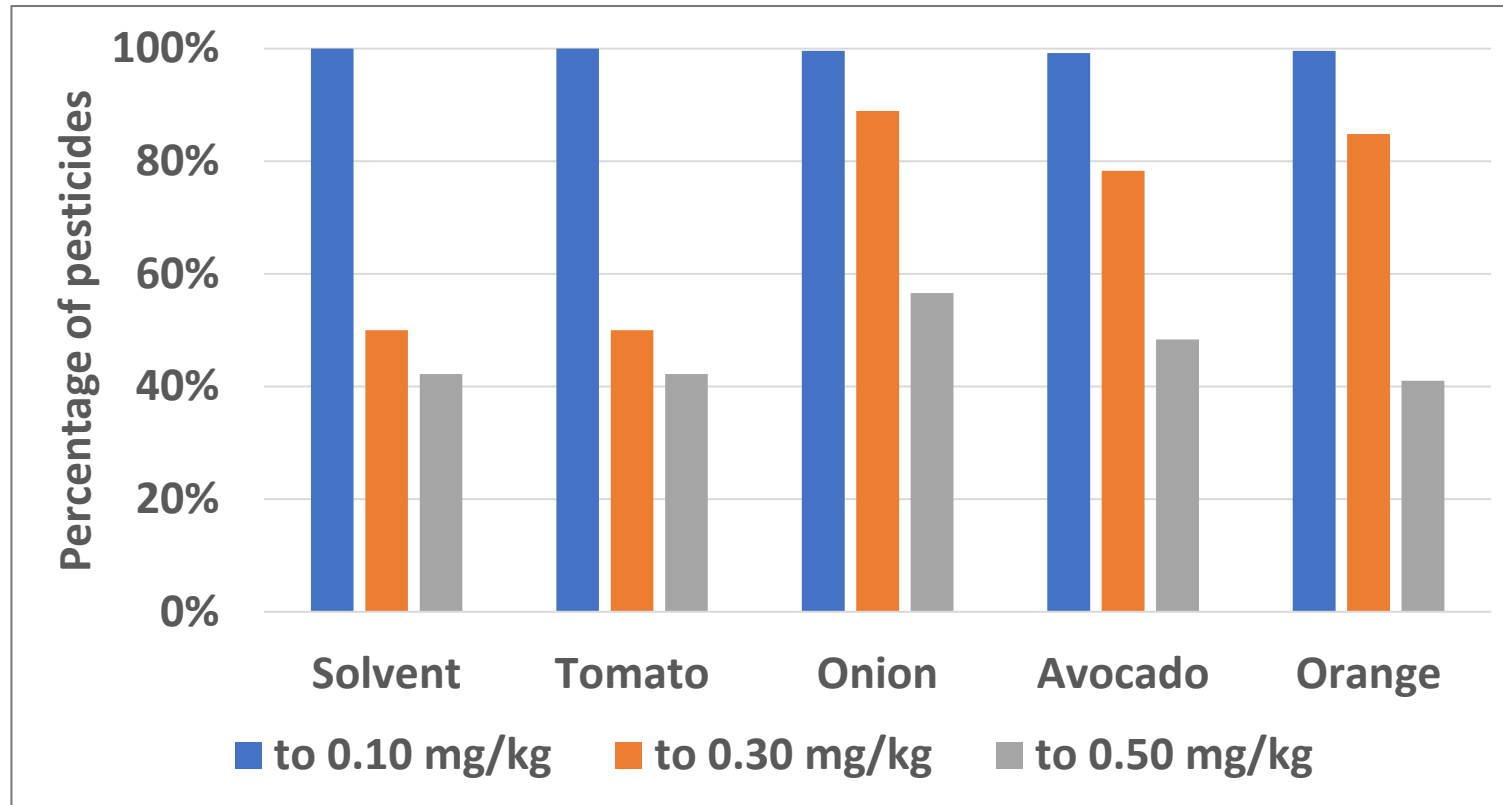
Target pesticides- validation



244 pesticides

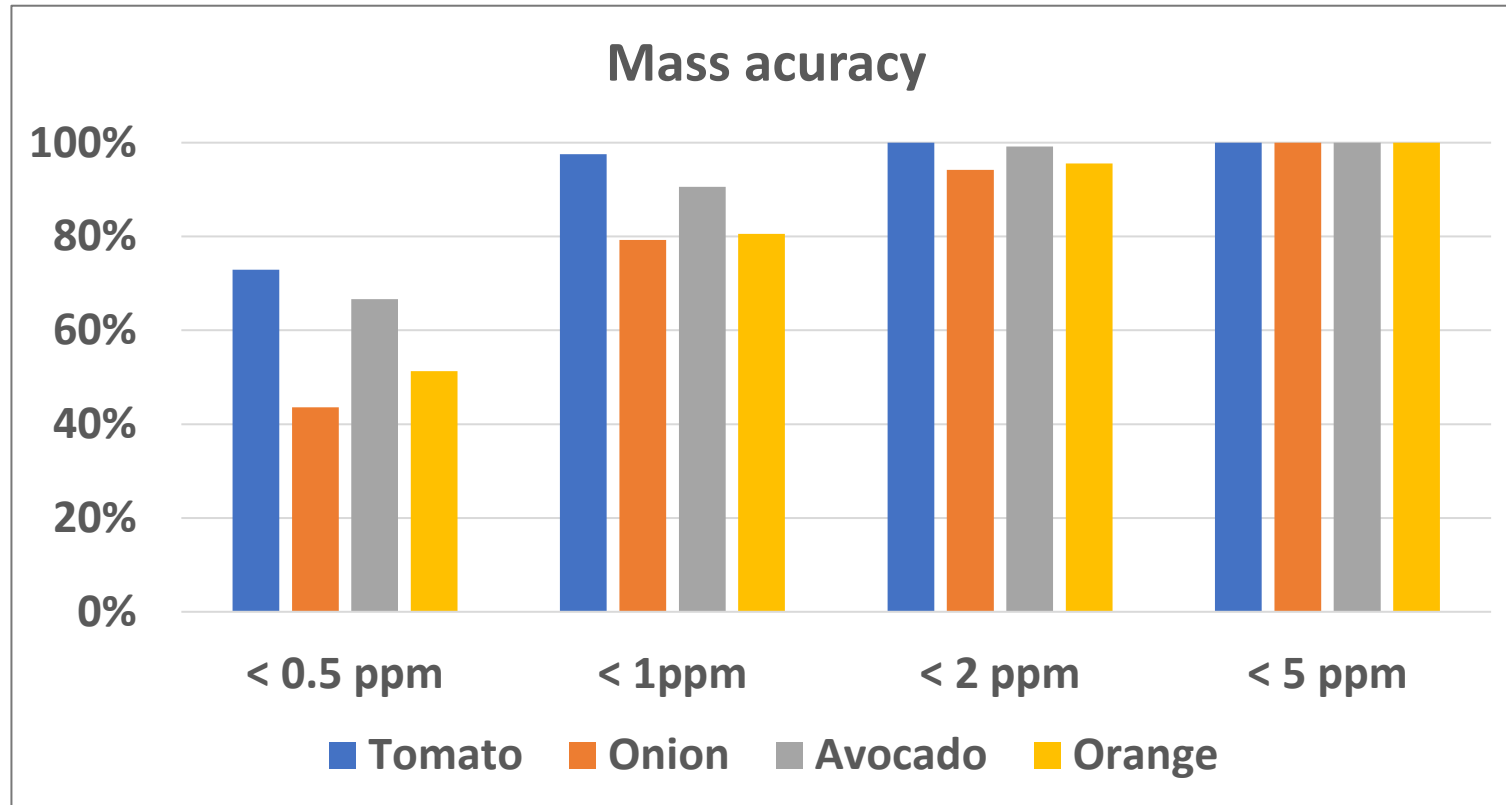


Linearity

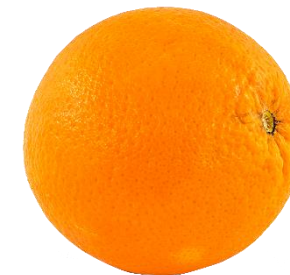


244 pesticides

Mass accuracy



0.01 mg/kg



Real samples

Target list 244 pesticides

Orange	23
Onion	15
Kiwi	9
Strawberry	1
Pinapple	1
Rasisns	1
sum	50



19 blancs

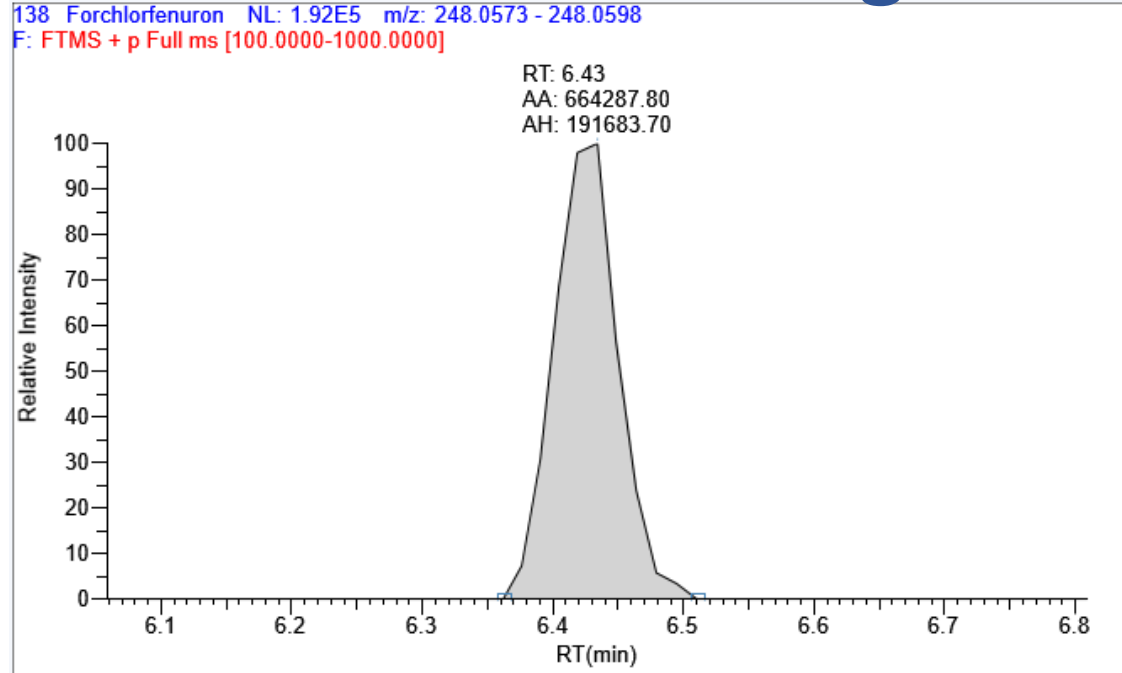
Imazalil	12	Metalaxyl	1
Acetamiprid	9	Mandipropamid	1
Pyrimethanil	6	Indoxacarb	1
Pyriproxyfen	5	Hexythiazox	1
Thiabendazole	3	Fluxapyroxad	1
Propiconazole	2	Fluopyram	1
Phosmet	2	Flubendiamide	1
Imidacloprid	2	Famoxadone	1
Fenpyroximate	2	Etofenprox	1
Boscalid	2	Proquinazid	1
Penconazole	1	Sulfoxaflor	1
Metrafenone	1	Tebuconazole	1
Methoxyfenozide	1	Zoxamide	1
		sum	61

Screening

Screening criteria:

- precursor ion mass tolerance 5 ppm (900 compounds in the data-base)
- isotopic pattern fit threshold 90%
- allowed mass deviation of isotopes 5 ppm
- allowed isotopic mass deviations of $\pm 10\%$
- library search score threshold 80
- library search passing value 80

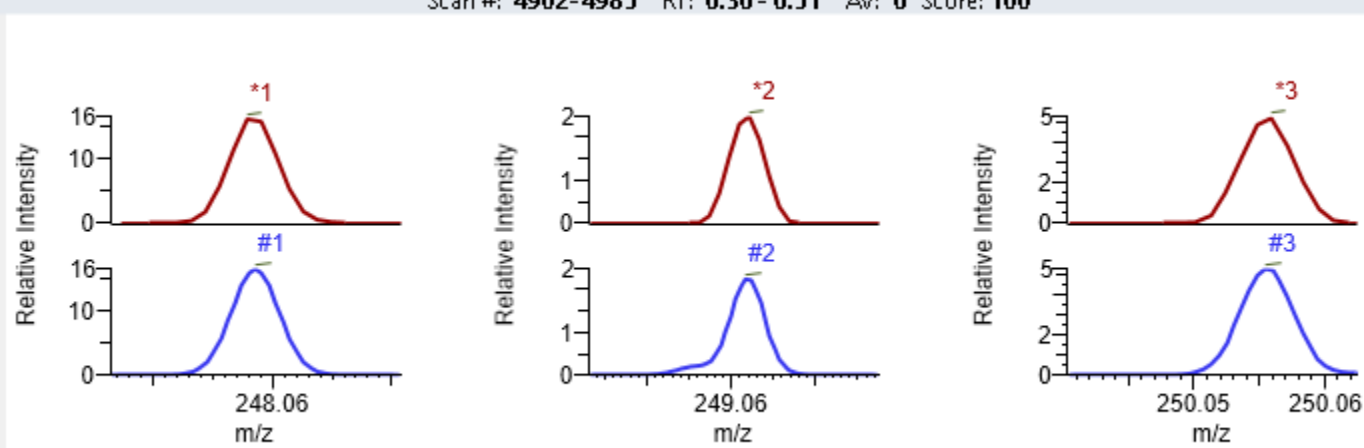
Screening



Full scan MS
 248.0585 ± 5ppm
 Mass error 0.17 ppm

Forchlorfenuron in kiwi sample

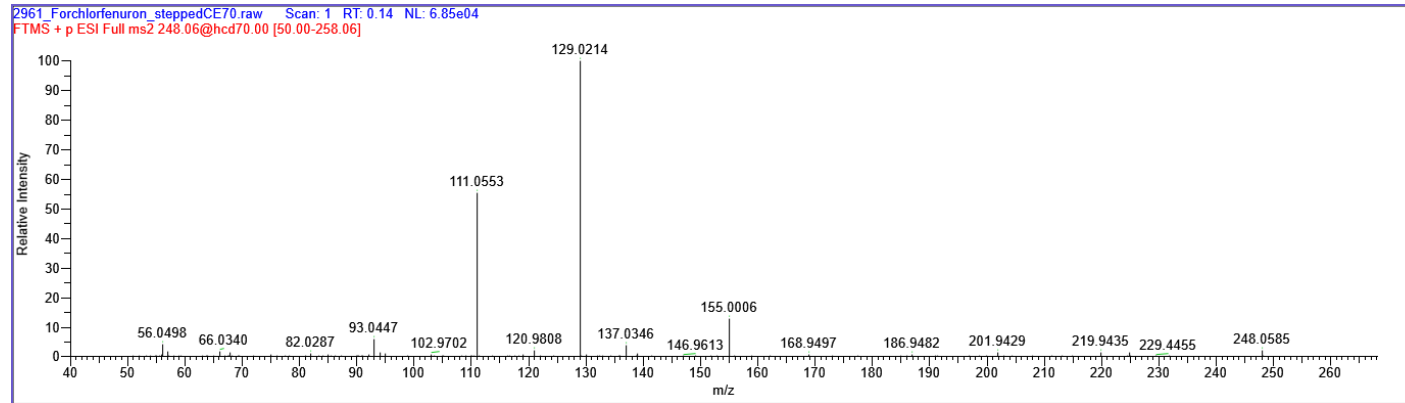
Scan #: 4902-4985 RT: 6.36 - 6.51 AV: 6 Score: 100



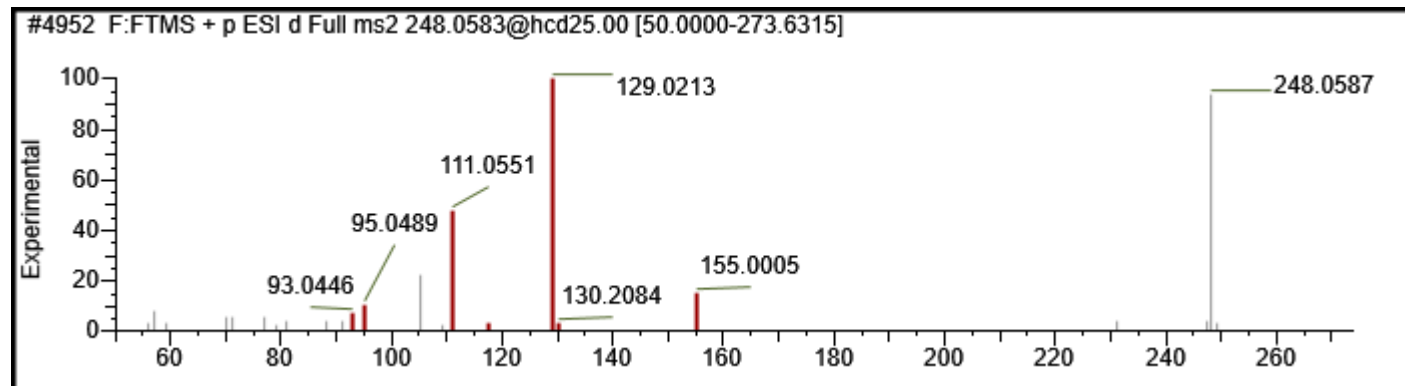
Detected isotopes

Expected isotopes

Screening



MS2 spectra (library)



MS2 spectra (sample)

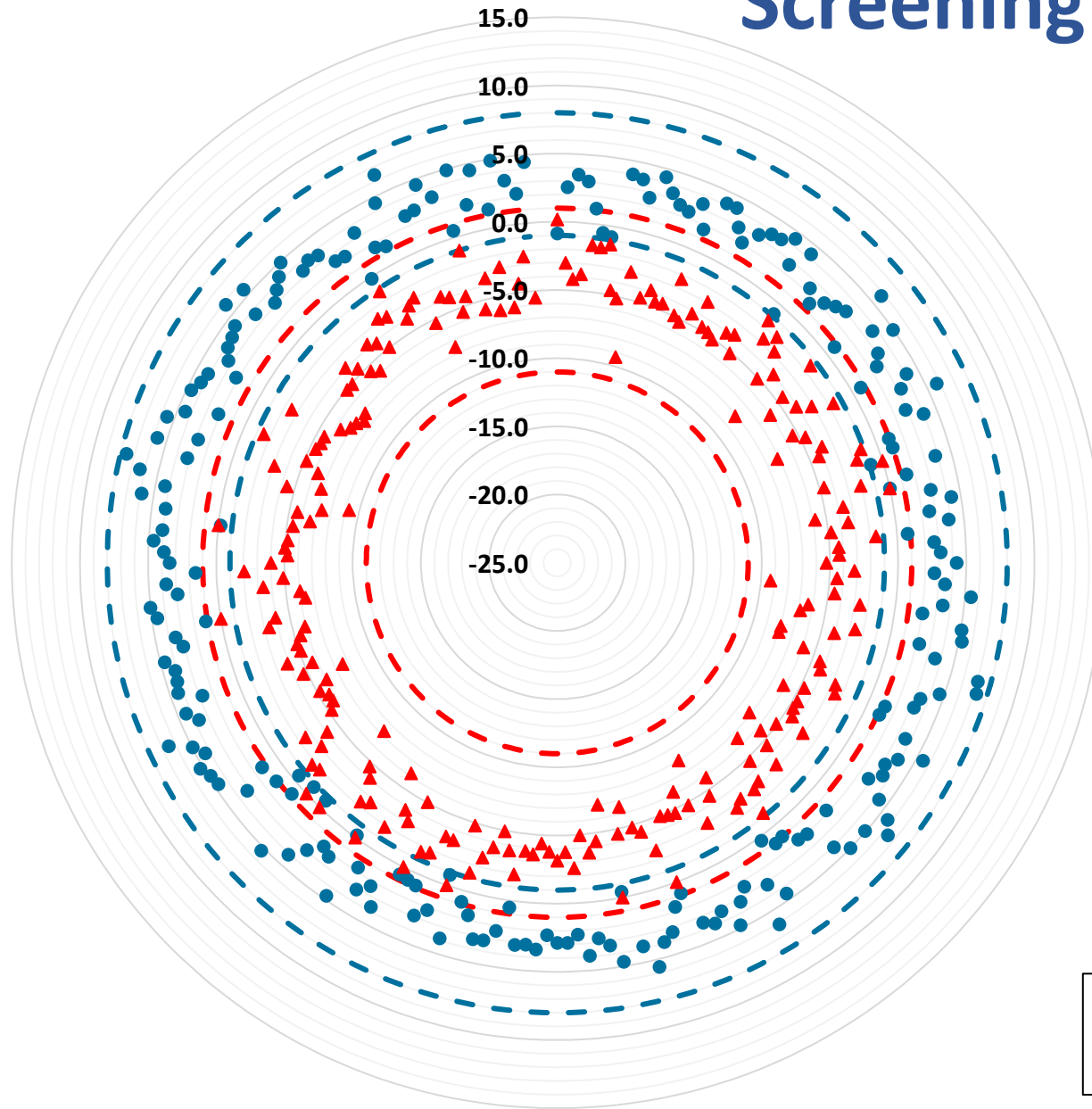
Forchlorfenuron in kiwi sample

Screening

Data-base with >900 compounds

Beauvericin	6
Imidacloprid,desnitro	4
Acetamiprid-metabolite-IM-2-1	2
Forchlorfenuron	2
Penicillic-Acid	2
Imidacloprid,desnitro-olefin	1
Imidacloprid,urea	1
Thiabendazole,5OH	1
sum	19

Screening



- log P
- - Min log P
- - Max log P
- ▲ log S
- - Min log S
- - Max log S

- $P = K_{ow}$ (pH = 7, T = 25 °C)
 - S = Water solubility (mol/L)

SUMMARY

The image features the letters 'EUR' in a bold, three-dimensional, light blue font. The letters are set against a solid blue background. Surrounding the text are several faint, light blue five-pointed stars, arranged in a circular pattern similar to the European Union flag. The overall aesthetic is clean and modern, with a slight shadow effect on the letters to give them depth.

EUR