Mass spectrometry: major improvements and new concepts in food analysis

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Overview

- Ion mobility-mass spectrometry IMS-MS
- Application to small molecules:
  - Screening of pesticides
  - Dioxin and PCBs
  - Peptides
  - Selenometabolites, metabolomics

- Molecular Imaging with MS

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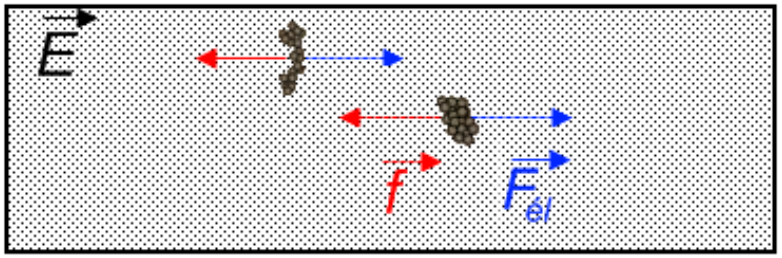
Basics: the linear drift tube

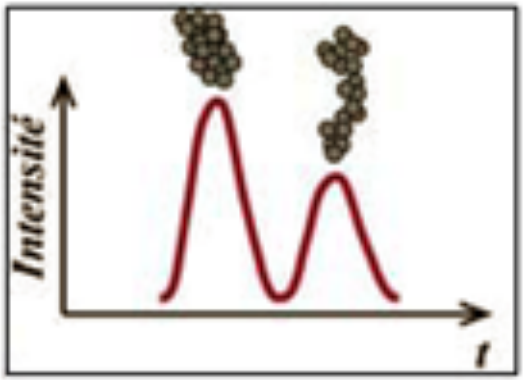
Drift tube



Very similar to electrophoresis

- Electric field→ Force $F_{él}$
- collisions → friction ***f***

- ***At force = 0,*** stationnary velocity v_d
- Ion Mobility in the gas phase $v_d = K.E$



- The method separate ions according to their shape (collision cross section)

$$K = \frac{3.e}{16N_0}\left(\frac{2\pi}{\mu k_B T}\right)^{1/2}\frac{1}{\Omega}$$

http://bowers.chem.ucsb.edu/

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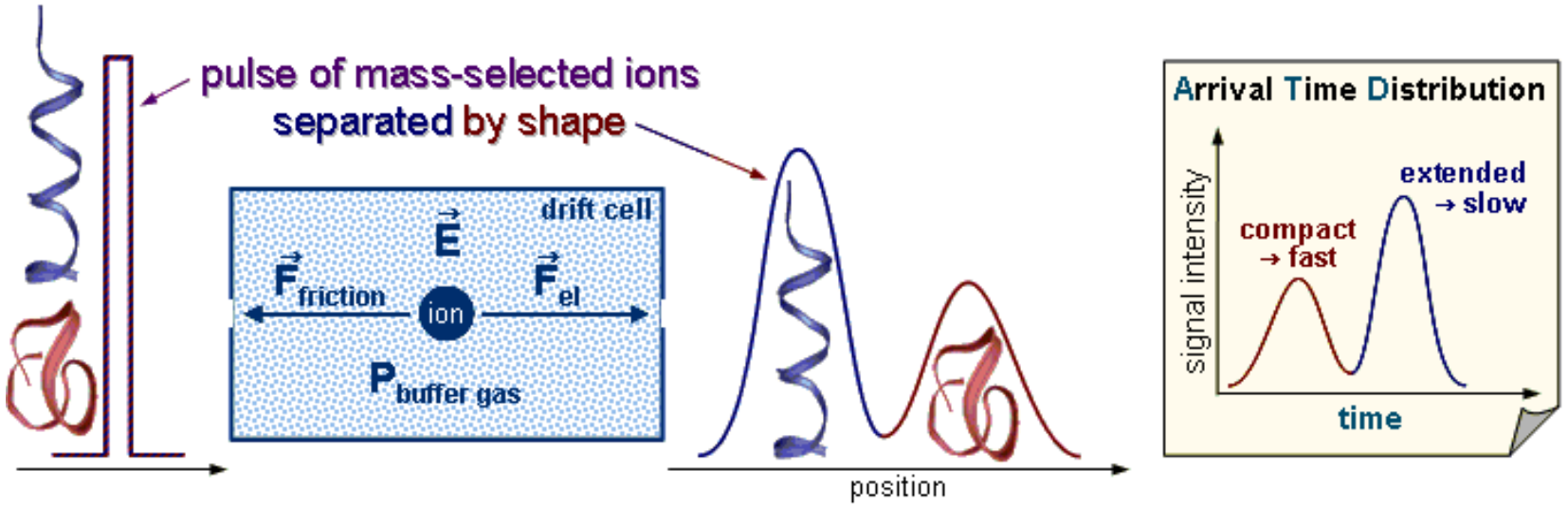
Ion mobility

Drift tube



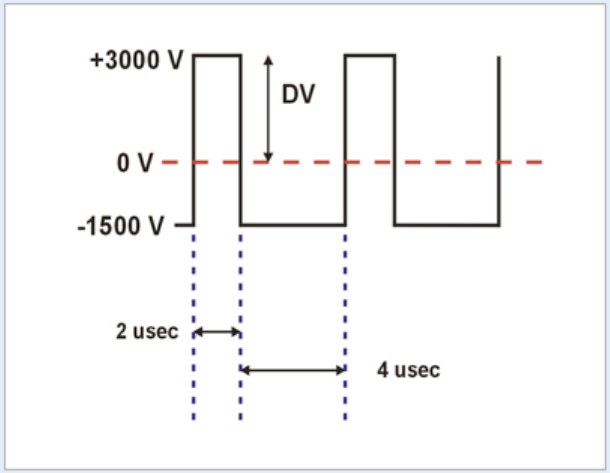
Mobility regimes

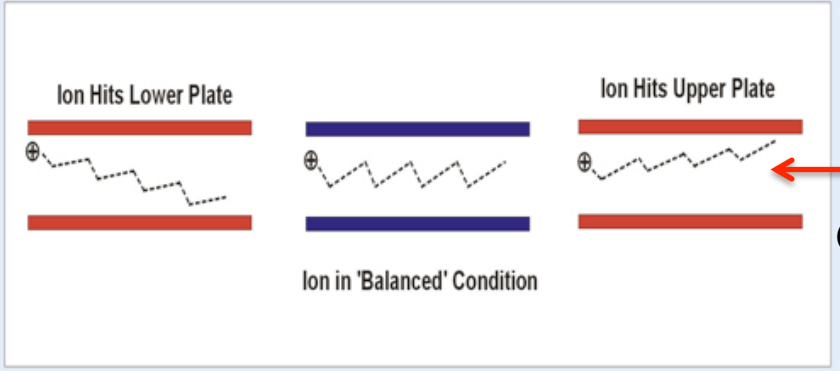
- High field
- Low field

- Side effects: heating up the ions

FAIMS (High-Field Asymmetric Waveform Ion Mobility Spectrometry)

- Separation of ions at high pressure and room temperature
- High E field (10 kV/cm) and tunable compensation field
- Asymetric field according to polarity
- Mobility proportional to electric field





http://www.faims.com

Mostly used as a prefilter

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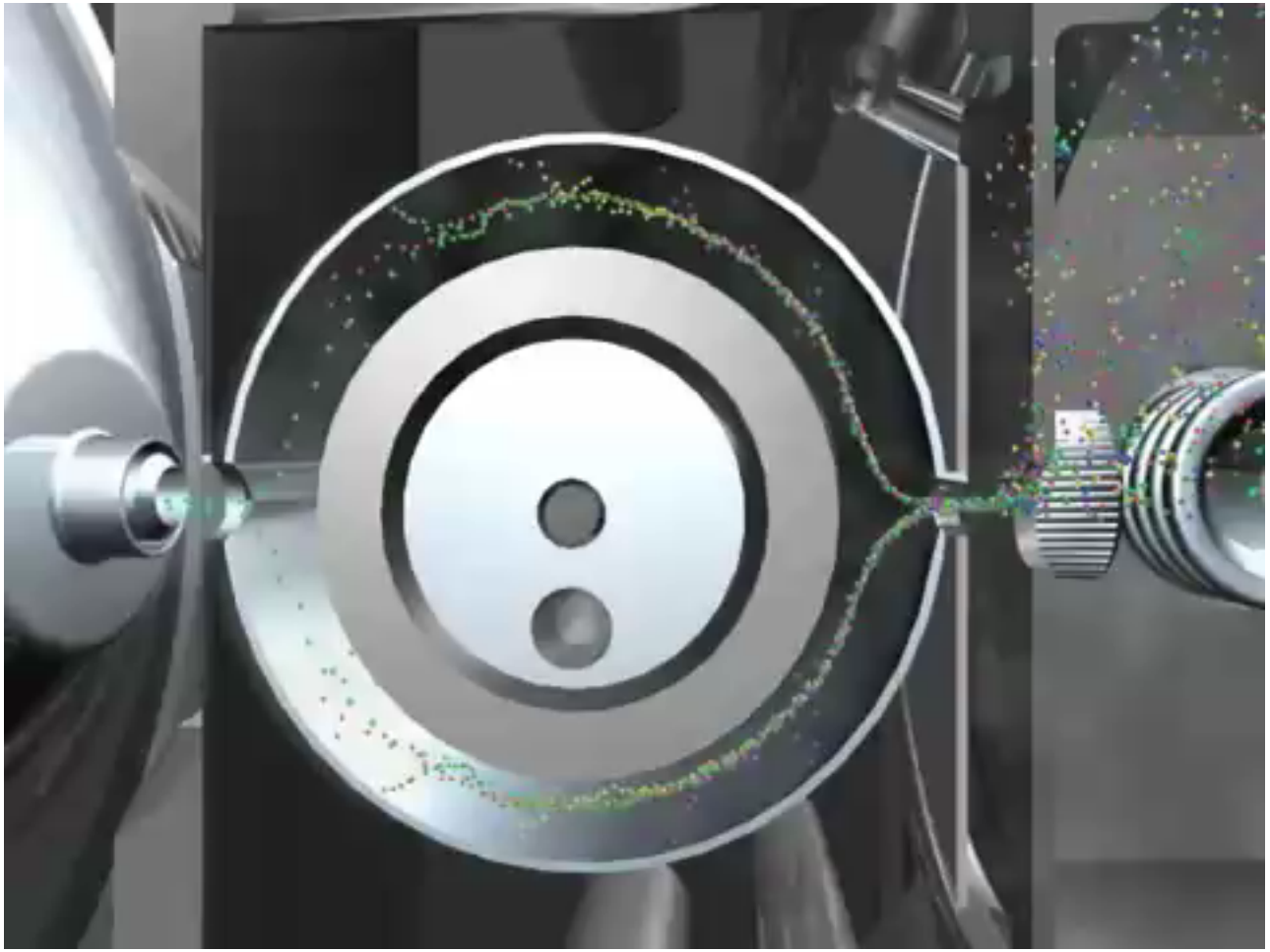
Illustration FAIMS



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Drift tube

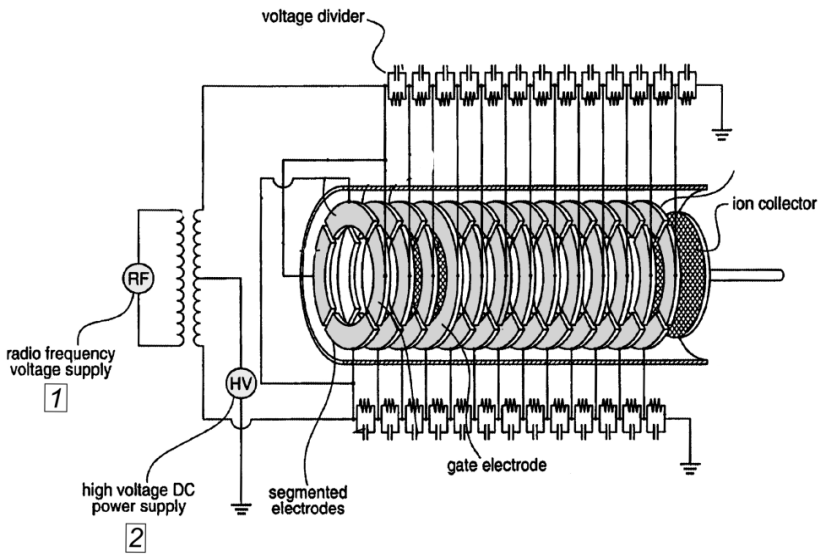
- Weak field
- Long drift tube
- Absolute cross sections
- Ions « Temperature » problem

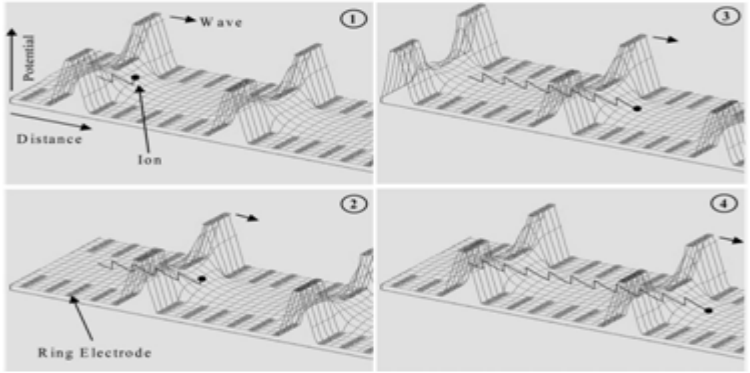
T-Wave ion Guide (T-Wave)

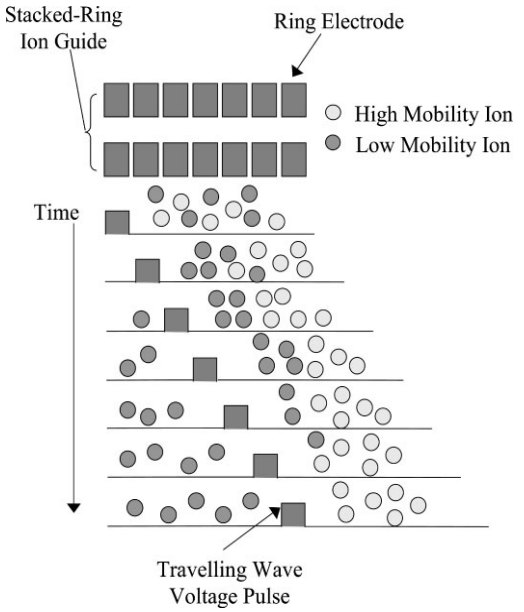
Series of circular electrodes between which a potential waves circulate

The system has the following effects:

- Focusing the ions (lateral potential)
- Inducing separation in the propagation direction according to the collisional cross section







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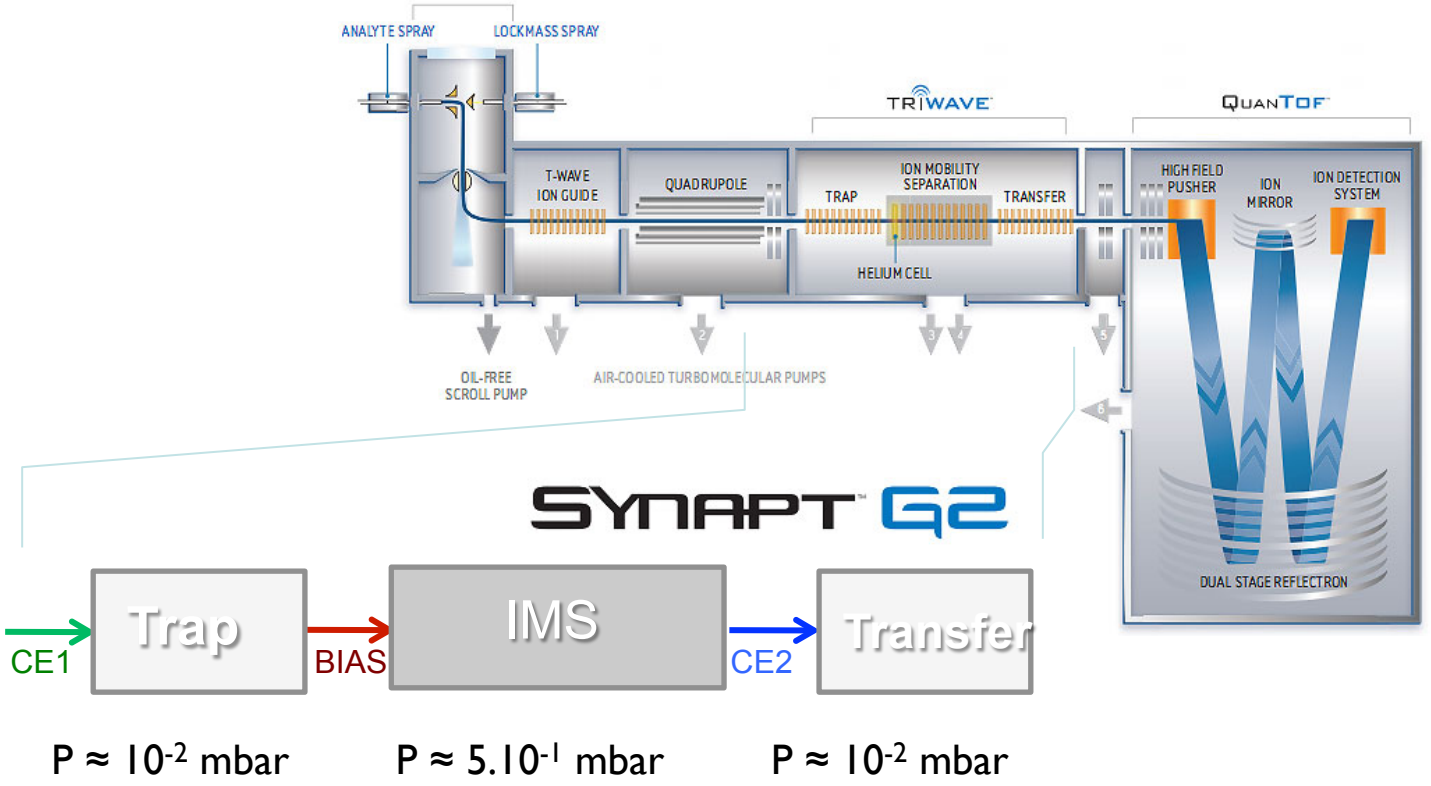
The Q-TOF type instrument (Synapt G2)



$P \approx 10^{-2}$ mbar $P \approx 5.10^{-1}$ mbar $P \approx 10^{-2}$ mbar

- ***Potential difference CE1***
- ***Potential difference Bias***
- ***Potential difference CE2***

within IM

Wave Amplitude
Wave velocity
Collision gas
Pressure of the collision gas

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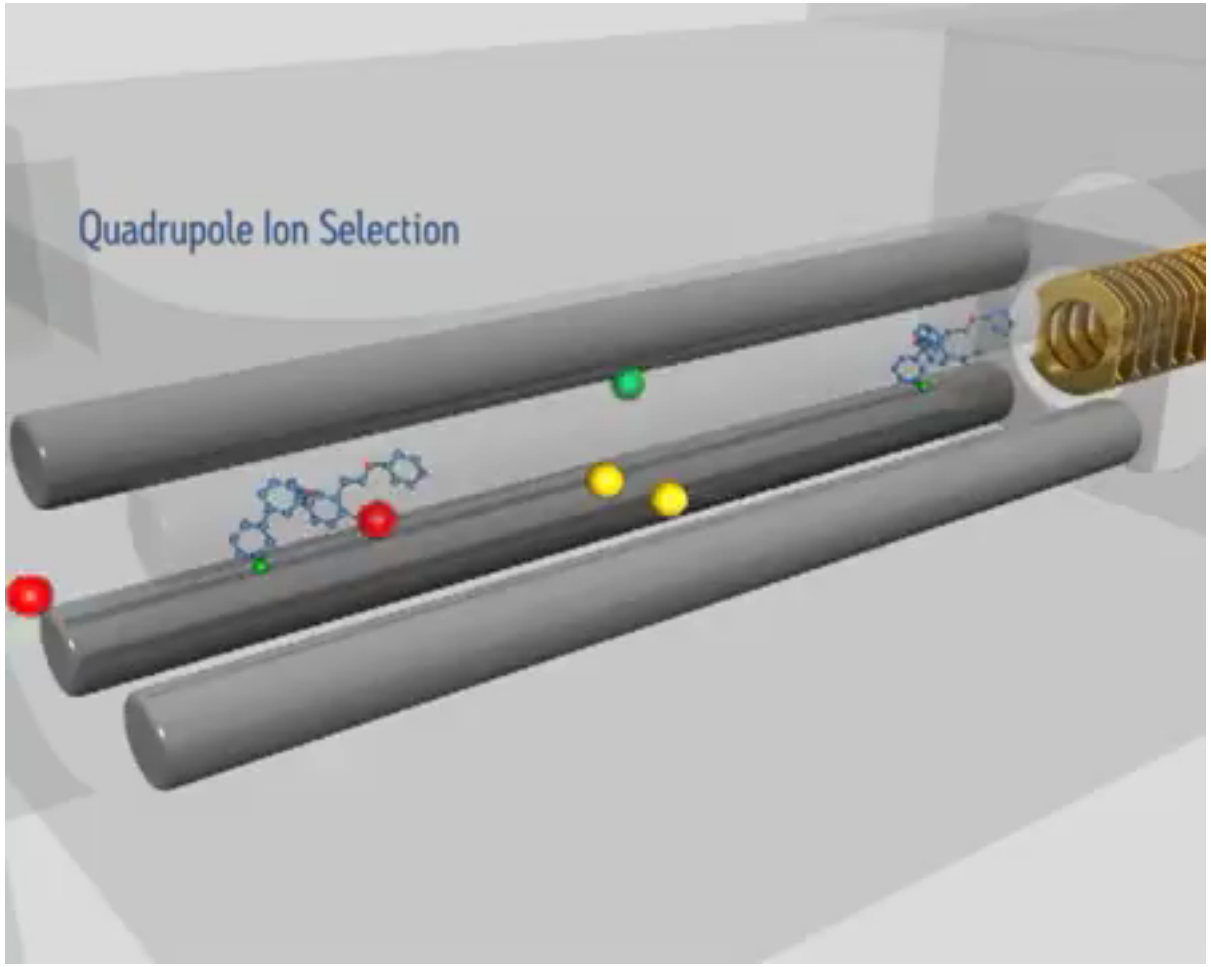
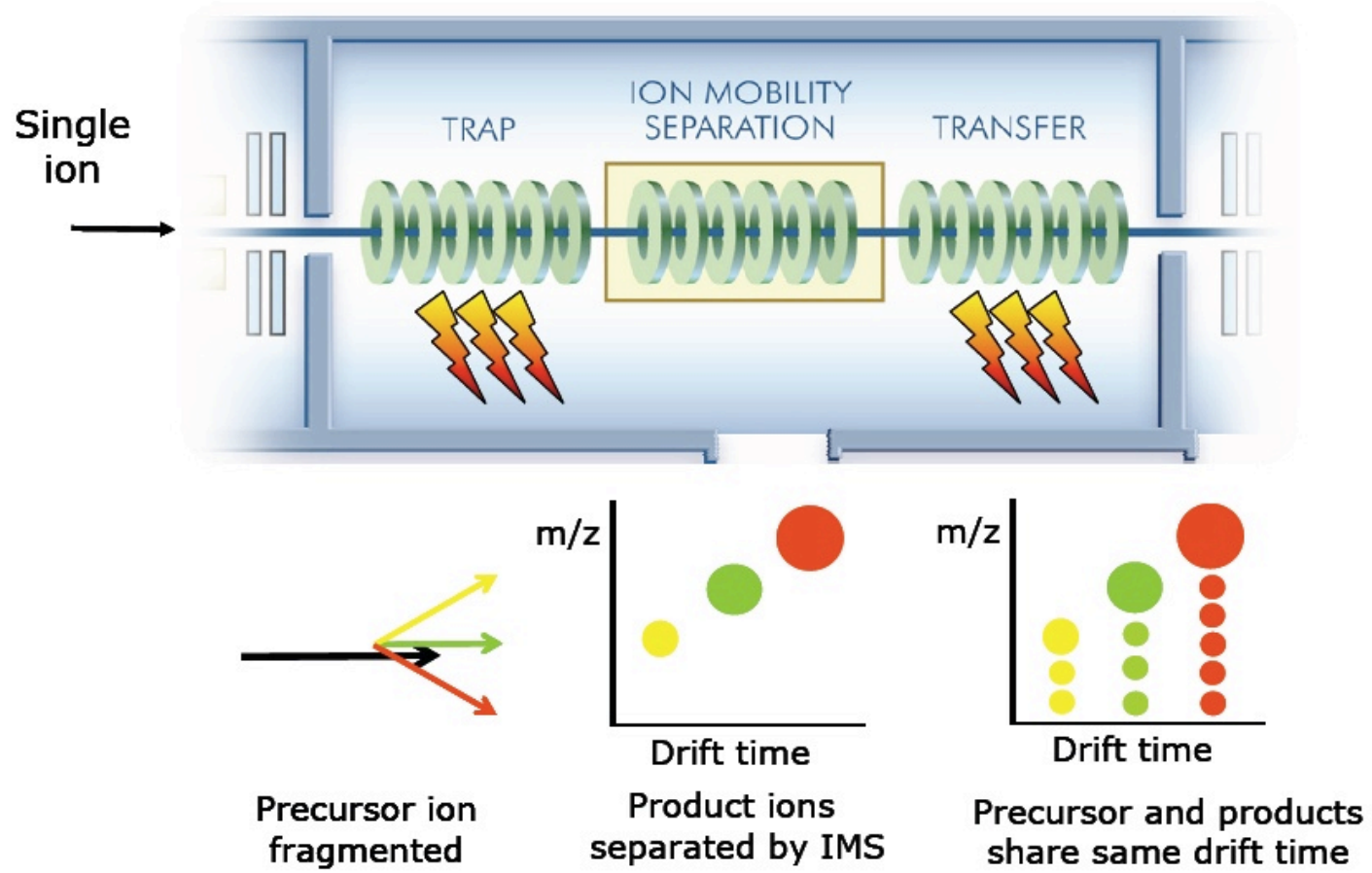
Illustration TWIG Waters



MS/MS “on the flight”



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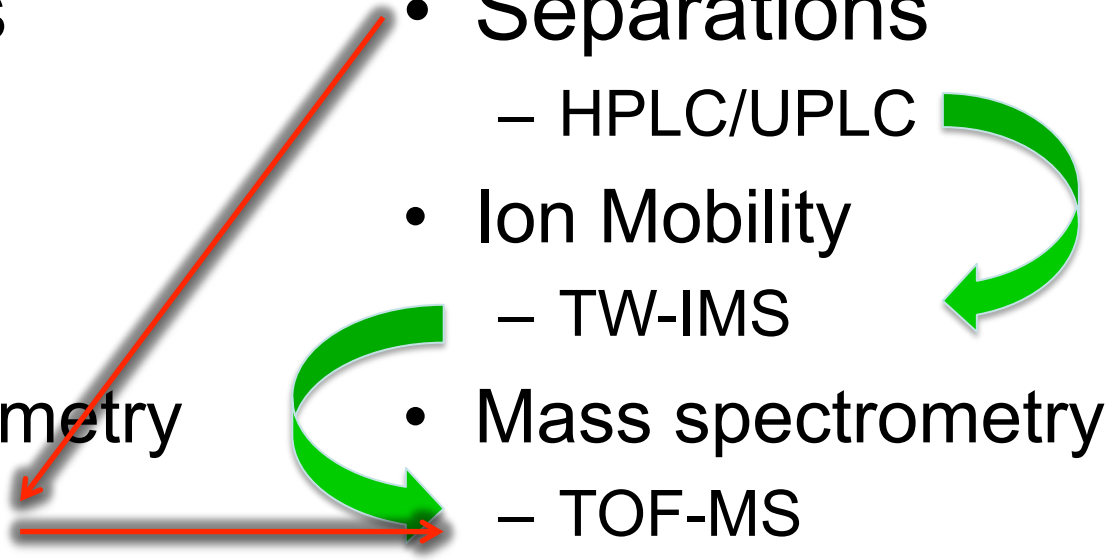
Non-dispersive vs dispersive process

Non dispersive (Filter)

- Separations
  - SPE
- Ion Mobility
  - FAIMS
- Mass spectrometry
  - Quadrupole
  - Magnetic sector

Dispersive

- Separations
  - HPLC/UPLC
- Ion Mobility
  - TW-IMS
- Mass spectrometry
  - TOF-MS
  - FT-ICR
  - FT-Orbi



HPLC (Seconds) >TW-IMS (mSeconds) > TOF-MS (µSeconds)

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Why integrate Chromatography + IMS + MS

‘*Since IMS separation is based on a different physical property from both HPLC/ GC and MS, it can provide significant benefits…*

…an additional separation which has advantages for the analysis of very complex mixtures’

C. Eckers *et al*.

Rapid Commun. Mass Spectrom. 2007, 21:1255

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Examples

- IMS large systems: proteins, DNA, polymers

- IMS small systems: amino acids, peptides, metabolites, pesticides, contaminants (dioxin and PCB), …

First example

Pesticide residues in food

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Identification criteria for pesticides (SANCO/10684)

Chromatography coupled to mass spectrometry:

- Retention time
- m/z
- Abundance data

Table 3 Identification requirements for different types of mass spectrometers

MS mode:	Single MS (standard mass resolution)	Single MS (high resolution/high mass accuracy)	MS/MS
Typical systems	Quadrupole, ion trap, time of flight (TOF)	TOF, Orbitrap, FTMS, magnetic sector	Triple quadrupole ion trap, hybride MS (e.g. Q-TOF
Acquisition:	Selected ion monitoring (SIM)	Selected ion monitoring (SIM)	(SRM/MRM), full scan product-ion spectra
Requirements for identification:	≥ 3 diagnostic ions, (preferably including quasi molecular ion)	≥ 2 diagnostic ions (preferably including the quasi molecular ion). Mass accuracy < 5 ppm. At least one fragment ion.	≥ 2 product ions
Ion ratio(s):	according to Table 4		

Table 4. Default recommended maximum permitted tolerances for relative ion intensities using a range of spectrometric techniques[2]

Relative intensity (% of base peak)	EI-GC-MS (relative)	CI-GC-MS, GC-MSn, LC-MS, LC-MSn (relative)
> 50 %	± 10 %	± 20 %
> 20 % to 50 %	± 15 %	± 25 %
> 10 % to 20 %	± 20 %	± 30 %
≤ 10%	± 50%	± 50 %

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Identification criteria for pesticides (SANCO/10684)

'…f*or a higher degree of confidence in identification…*

It can be achieved through

- *Additional MS information*
  - *Full scan spectra*
  - *Additional accurate mass (fragment) ions*
  - *Additional product ions*
  - *Accurate mass product ions*

- *Additional evidence may be sought using a different chromatographic separation system…*

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Screening of pesticides by HPLC-ESI-Q-IMS-TOF

Development and optimization of the method:

- Many IMS-MS parameters to tune and to optimize
- Experimental design approach (Placket Burmann, Central Composite Design)
- More details in S. Goscinny talk…

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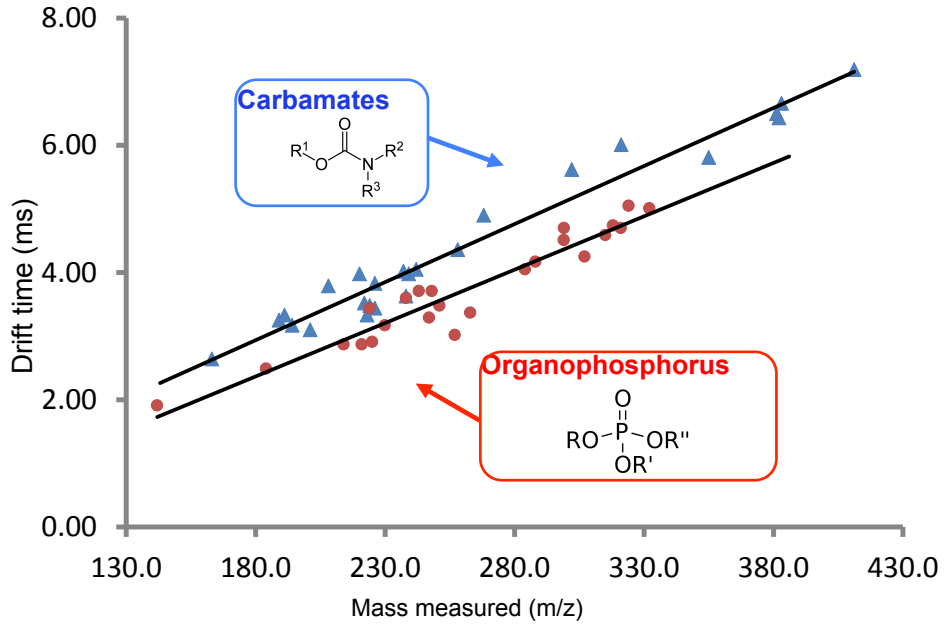
Screening of pesticides by HPLC-ESI-Q-IMS-TOF

- Discrimination between classes of pesticides by IMS



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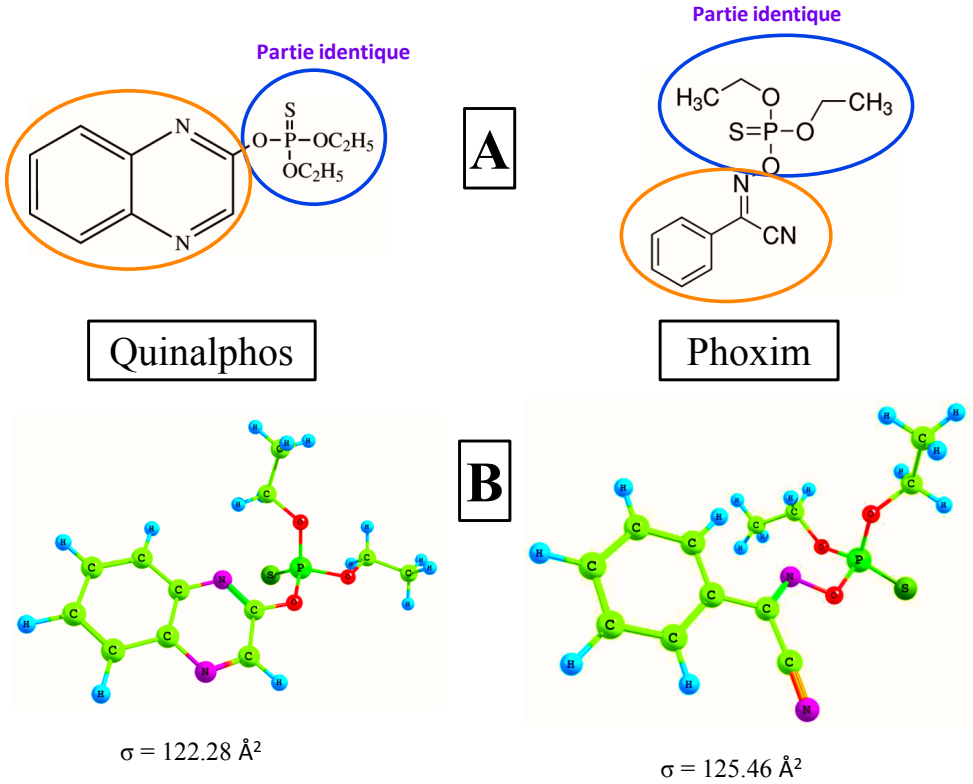
Screening of pesticides by HPLC-ESI-Q-IMS-TOF

- Discrimination between isobaric compounds coeluting in HPLC



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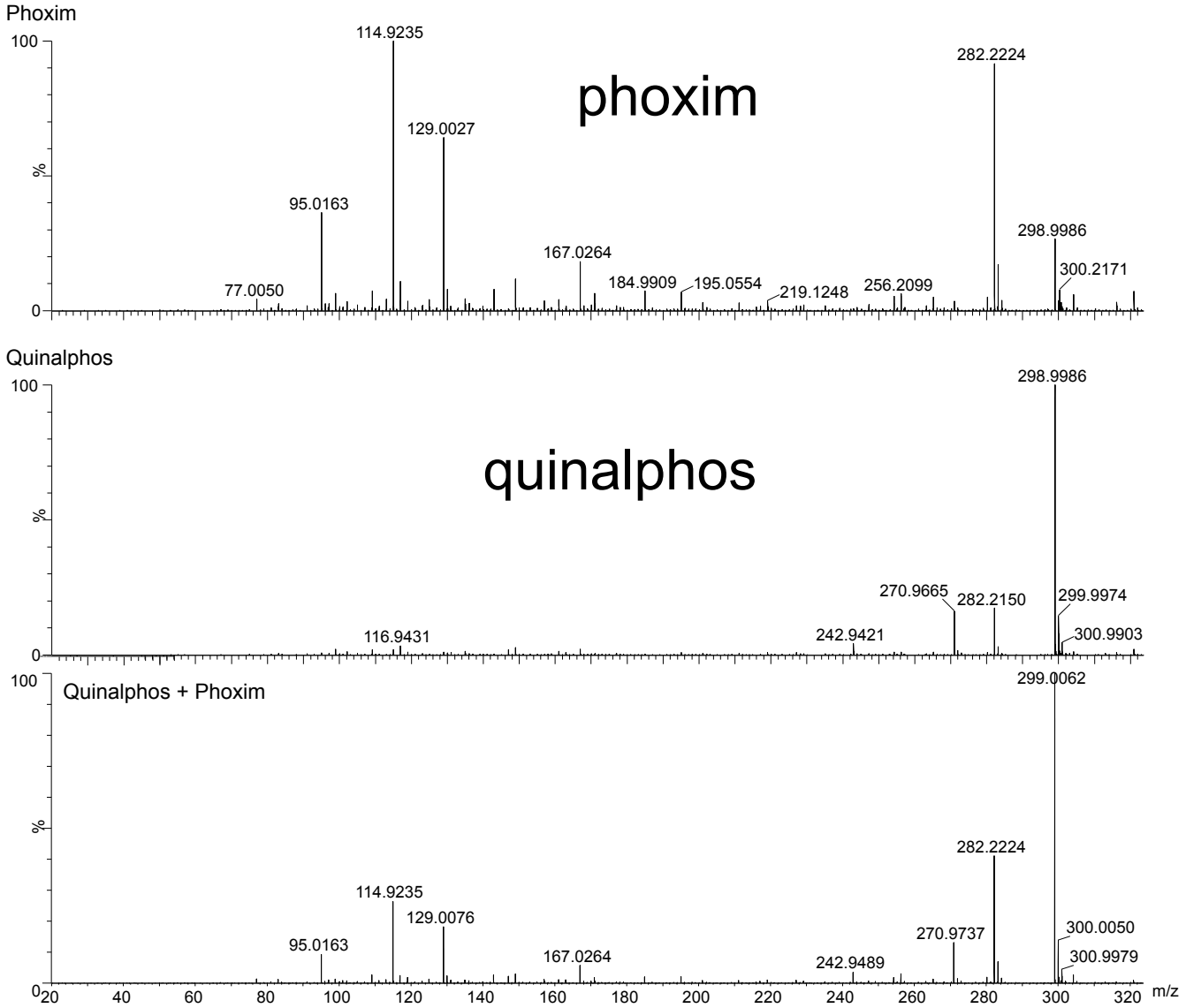
Equivalent ESI Q ToF MS1



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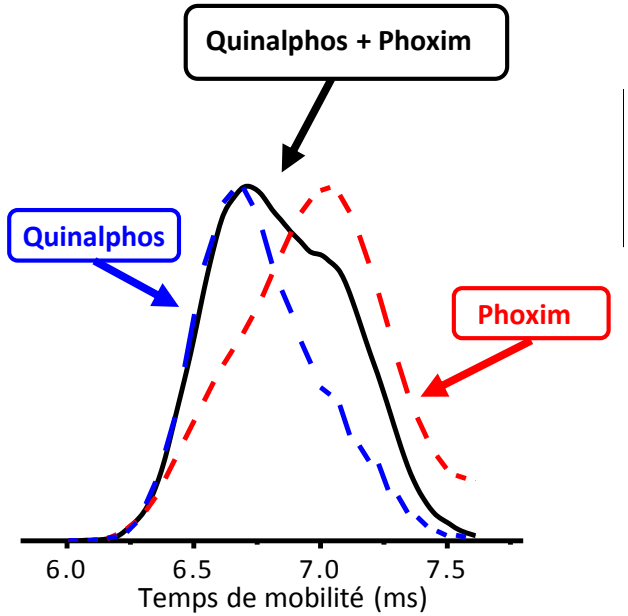
Screening of pesticides by HPLC-ESI-Q-IMS-TOF

Mobilogram of molecular ions



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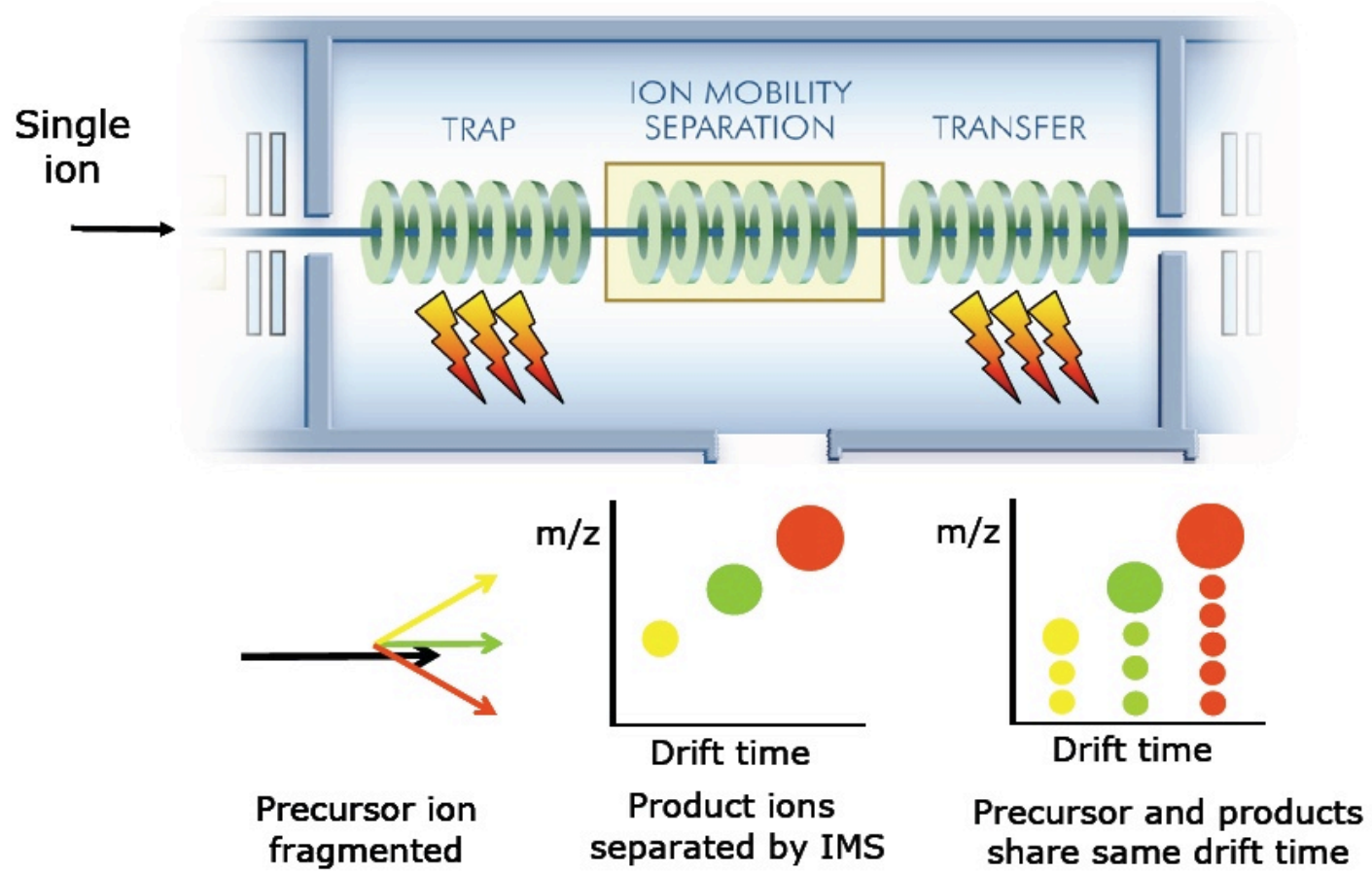
MS/MS “on the flight”



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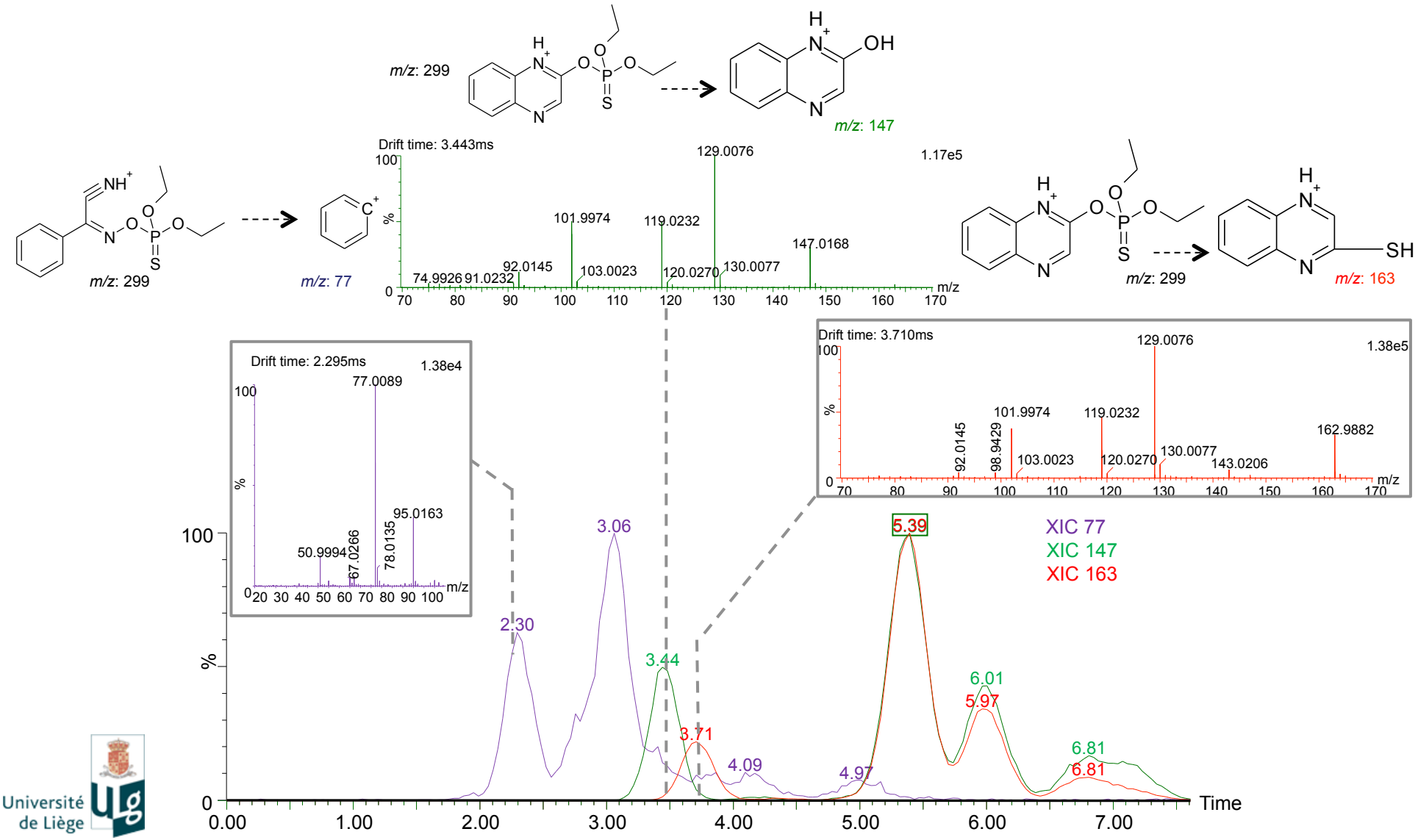
ESI – Trap MS/MS – IMS – Transfer MS/MS



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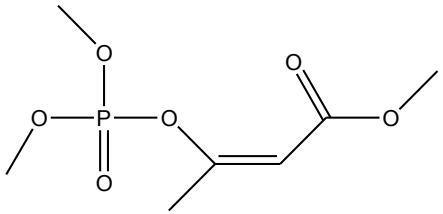
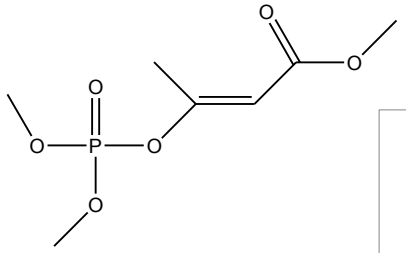
Screening of pesticides by HPLC-ESI-Q-IMS-TOF

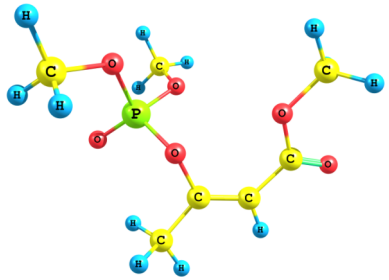
Discrimination between E:Z-isomers



(Z)-mevinphos

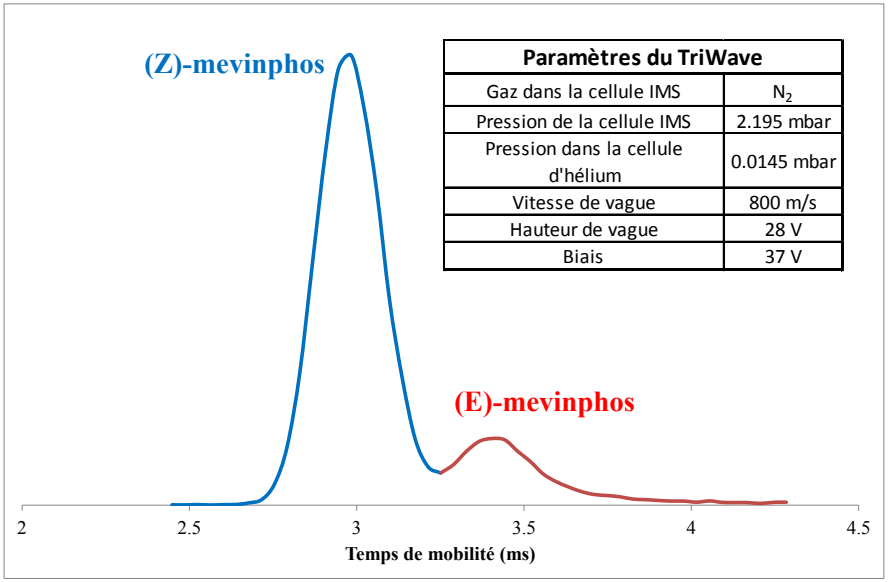


(E)-mevinphos



σ = 101.79 Å²

σ = 106.37 Å²



Paramètres du TriWave	
Gaz dans la cellule IMS	N_2
Pression de la cellule IMS	2.195 mbar
Pression dans la cellule d'hélium	0.0145 mbar
Vitesse de vague	800 m/s
Hauteur de vague	28 V
Biais	37 V

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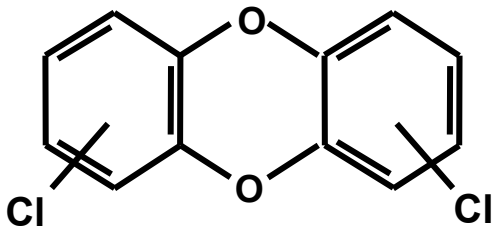
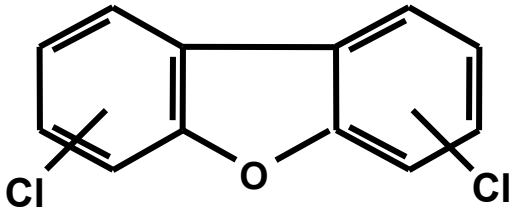
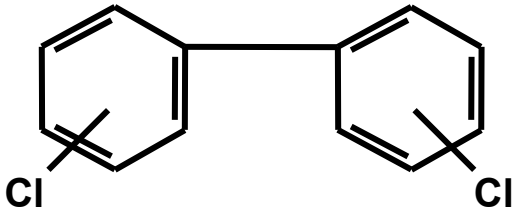
Second example

Halogenated aromatic compounds:

Dioxins and PCBs







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GC-EI-Q-IMS-TOF of Dioxin and PCBs

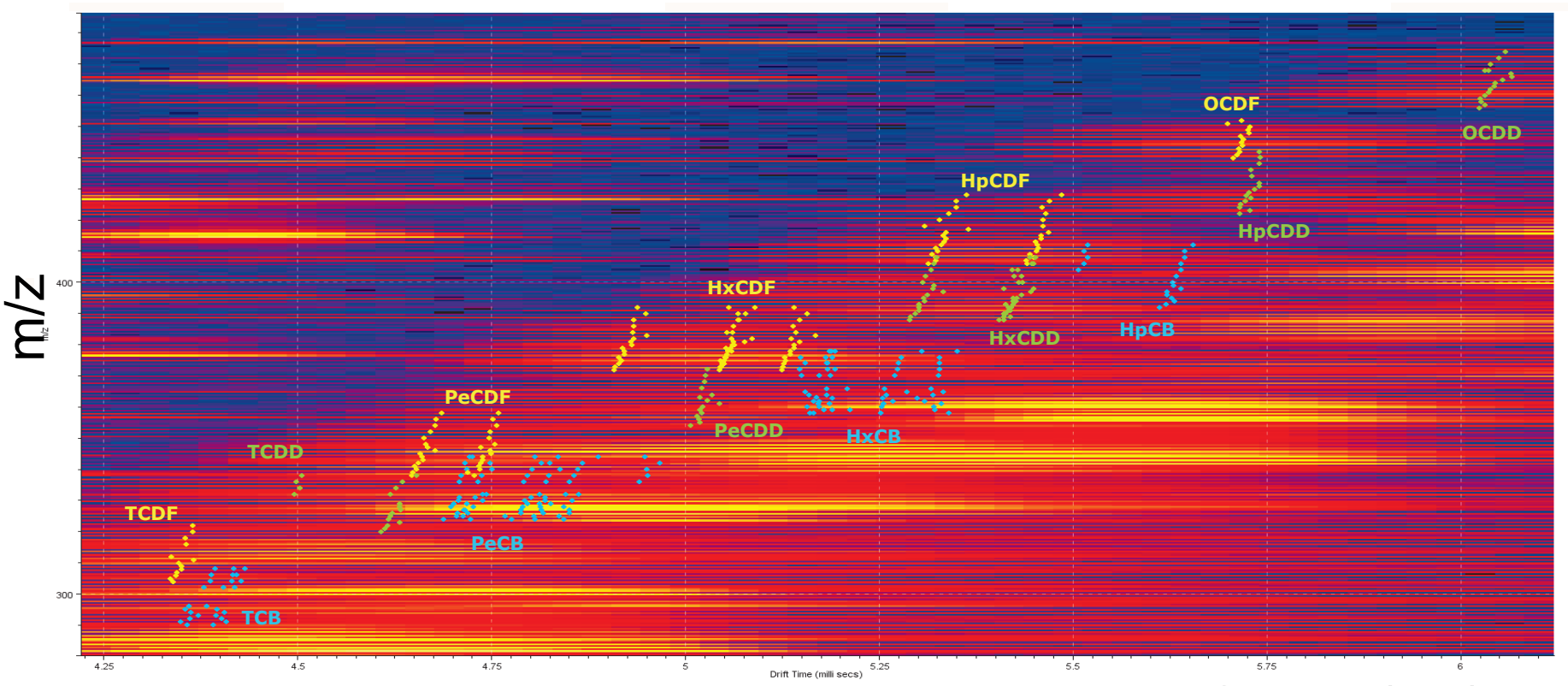

Figure 4. Ion mobility plot of dioxins, furans and biphenyls

Drift time (ms)

With the courtesy of Martin Green (Waters, UK)

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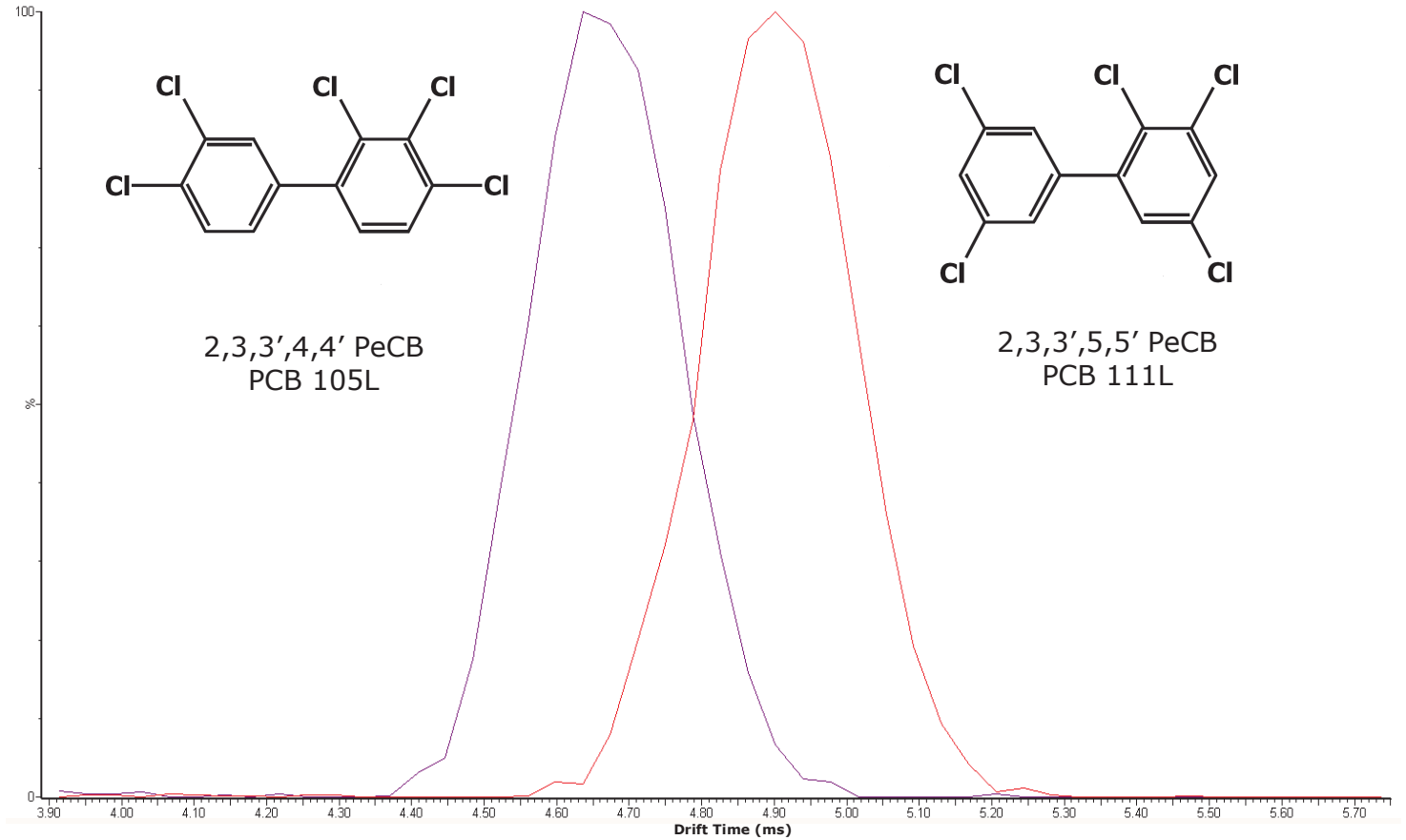
GC-EI-Q-IMS-TOF: Mobilogram of two PeCBs



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With the courtesy of Martin Green (Waters, UK)

GC-EI-Q-IMS-TOF of Dioxin and PCBs

Correlation between drift time and calculated cross section

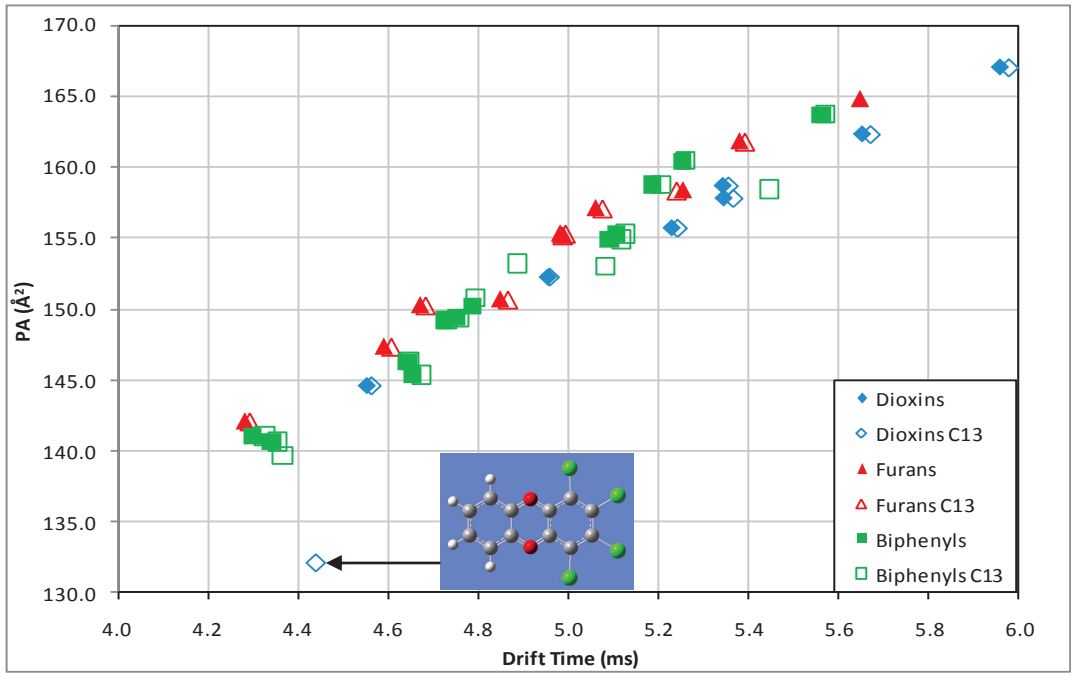

Figure 6. Drift time plotted against calculated cross-section

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With the courtesy of Martin Green (Waters, UK)

Third example

Peptides

(Venomics project)

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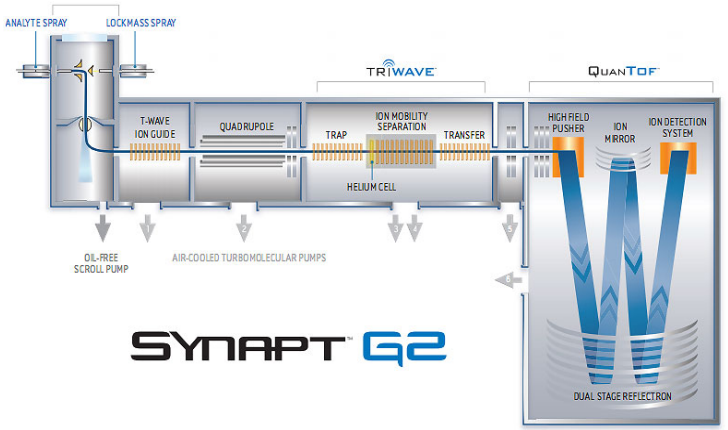
IMS of peptides (venomics project)

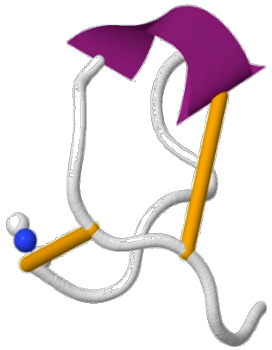
- Development of a robust disulfide bonds assignment method using ion Mobility Spectrometry (IMS):
  - "Gas phase electrophoresis"
  - Distinction of isobaric species based on their collision cross-section.



Proof-of-concept model:

- Partial reduction of α-conotoxin with a slight excess of TCEP (10 fold).

Conus consors



α-CnIa toxin (*Conus*)

GR**C**C**C**H PA**C**GK YYS**C** * Mass : 1541.6Da (oxidized)

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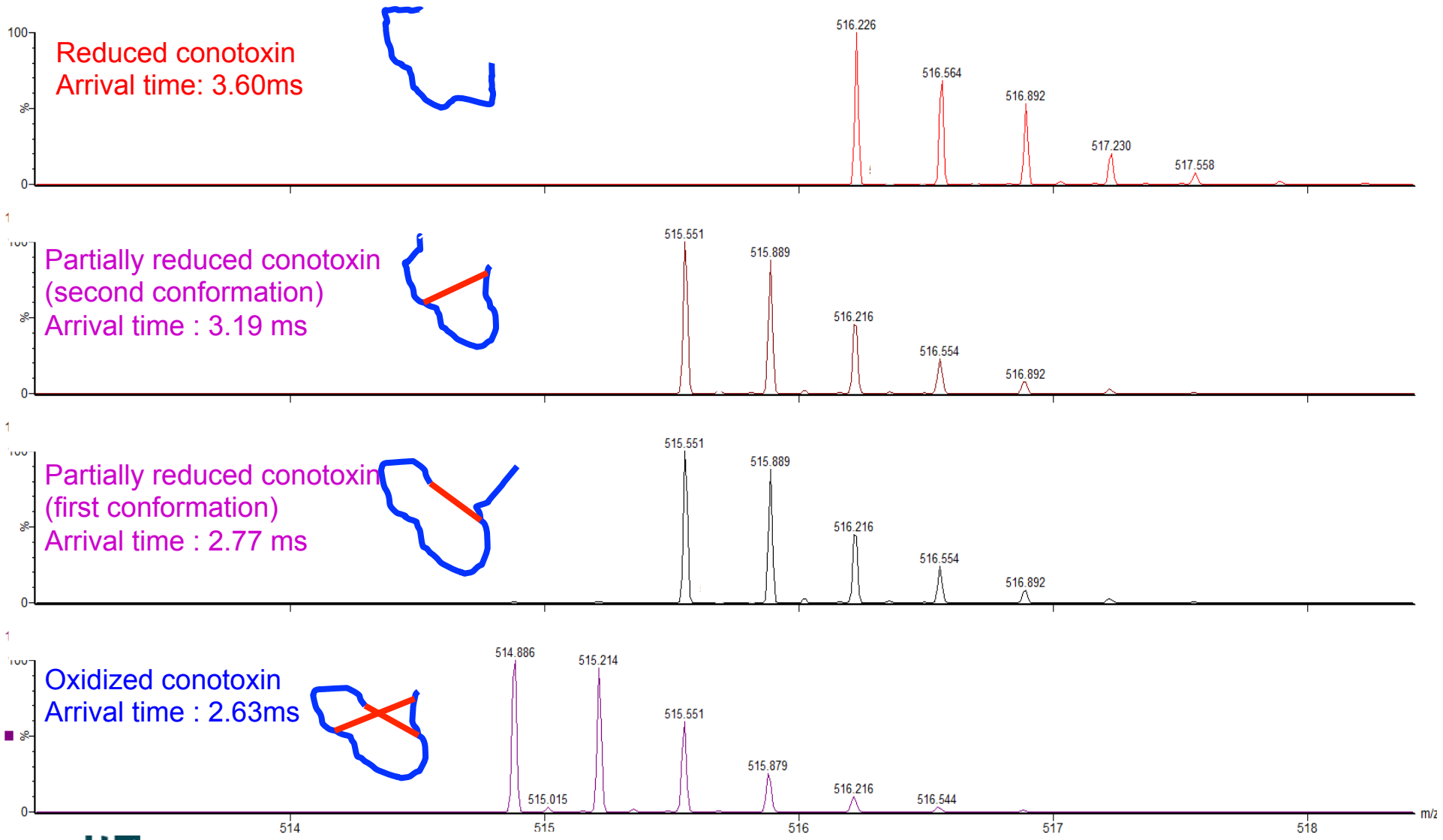
Mass Spectra extracted from the mobilogram (partially reduced toxin)



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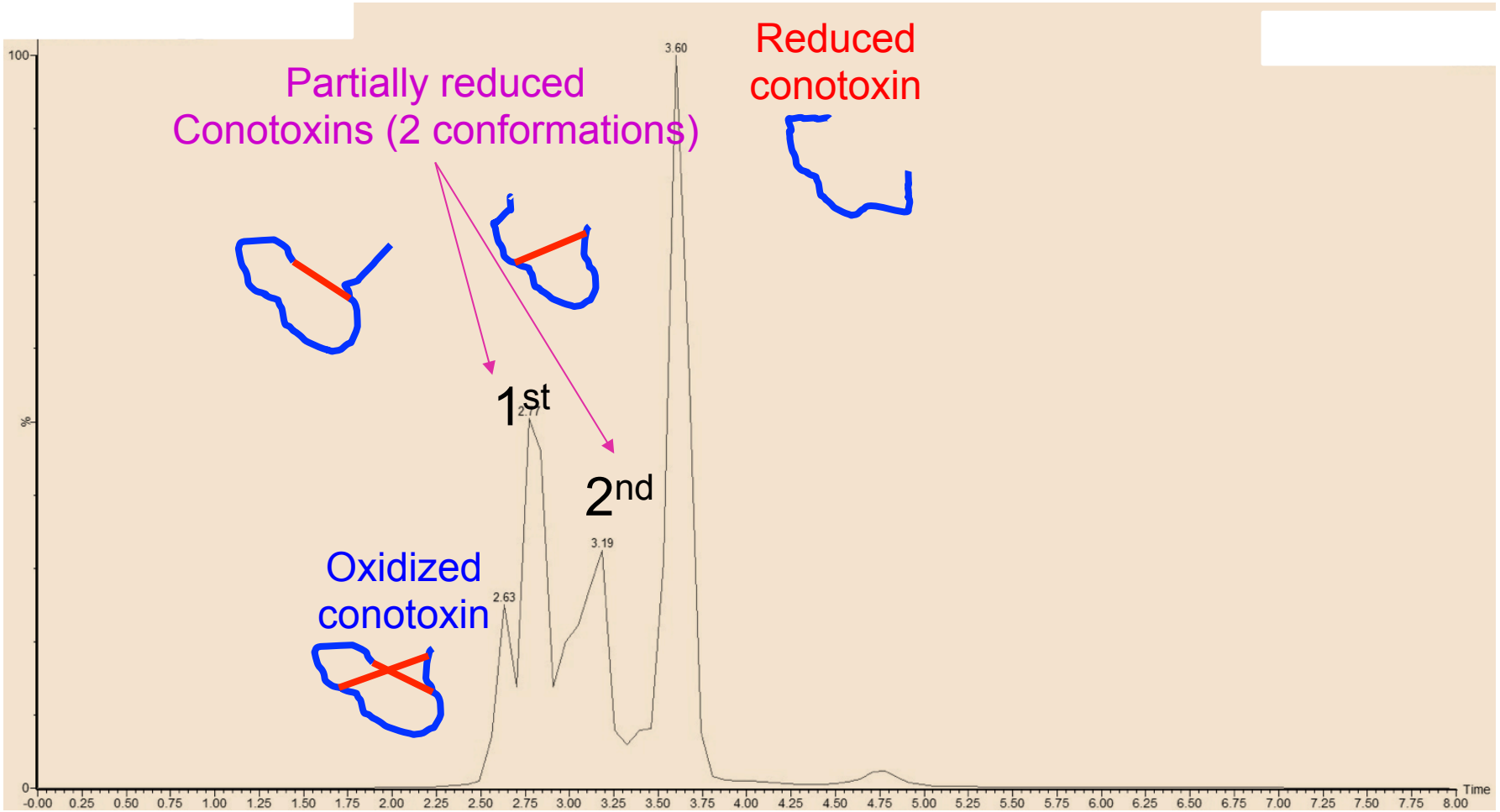
Four species can be seen in the mobilogram (partially reduced toxin)



3X charged 2X charged

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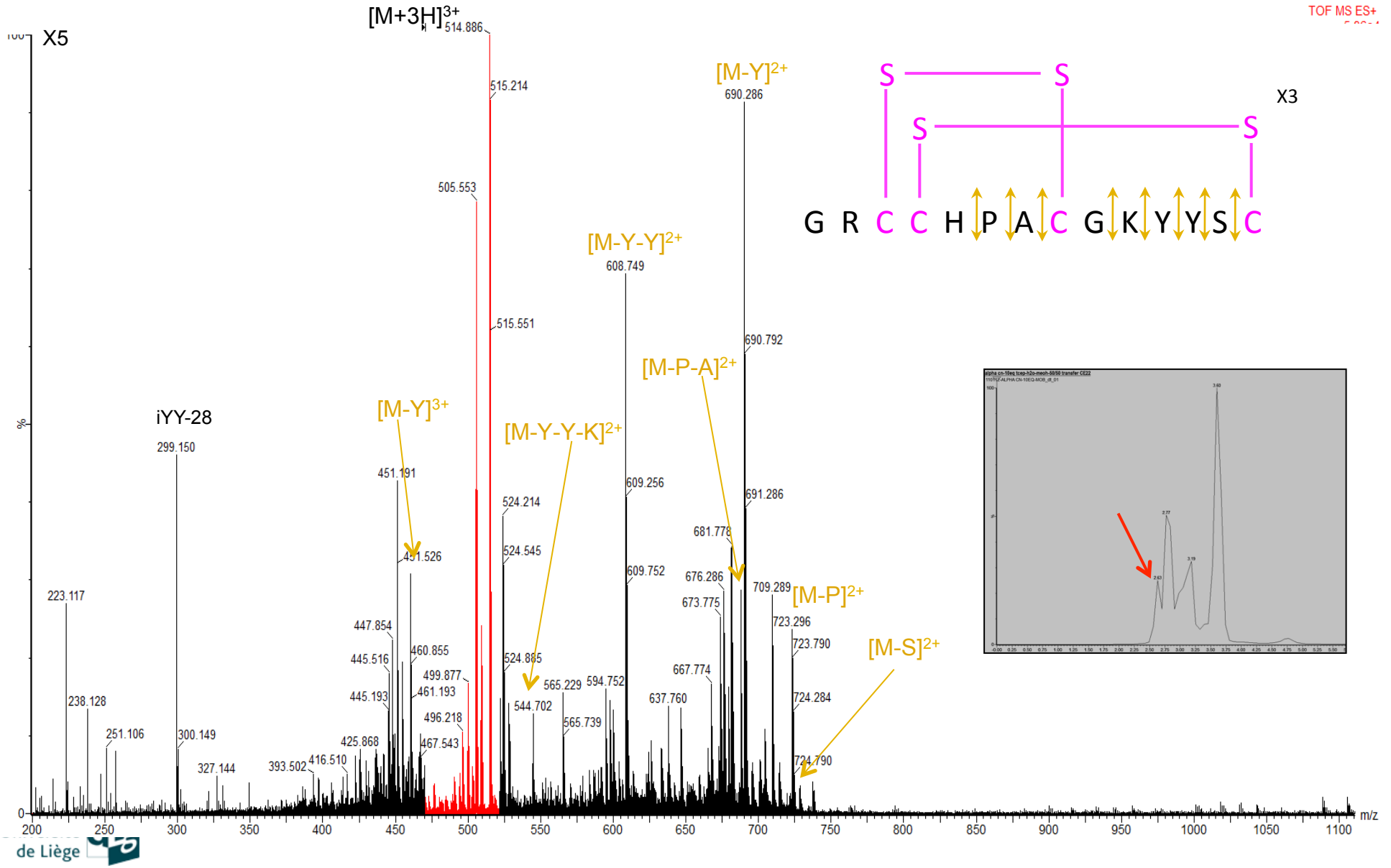
Fragmentation spectrum of the oxidized form (arrival time : 2.562ms)



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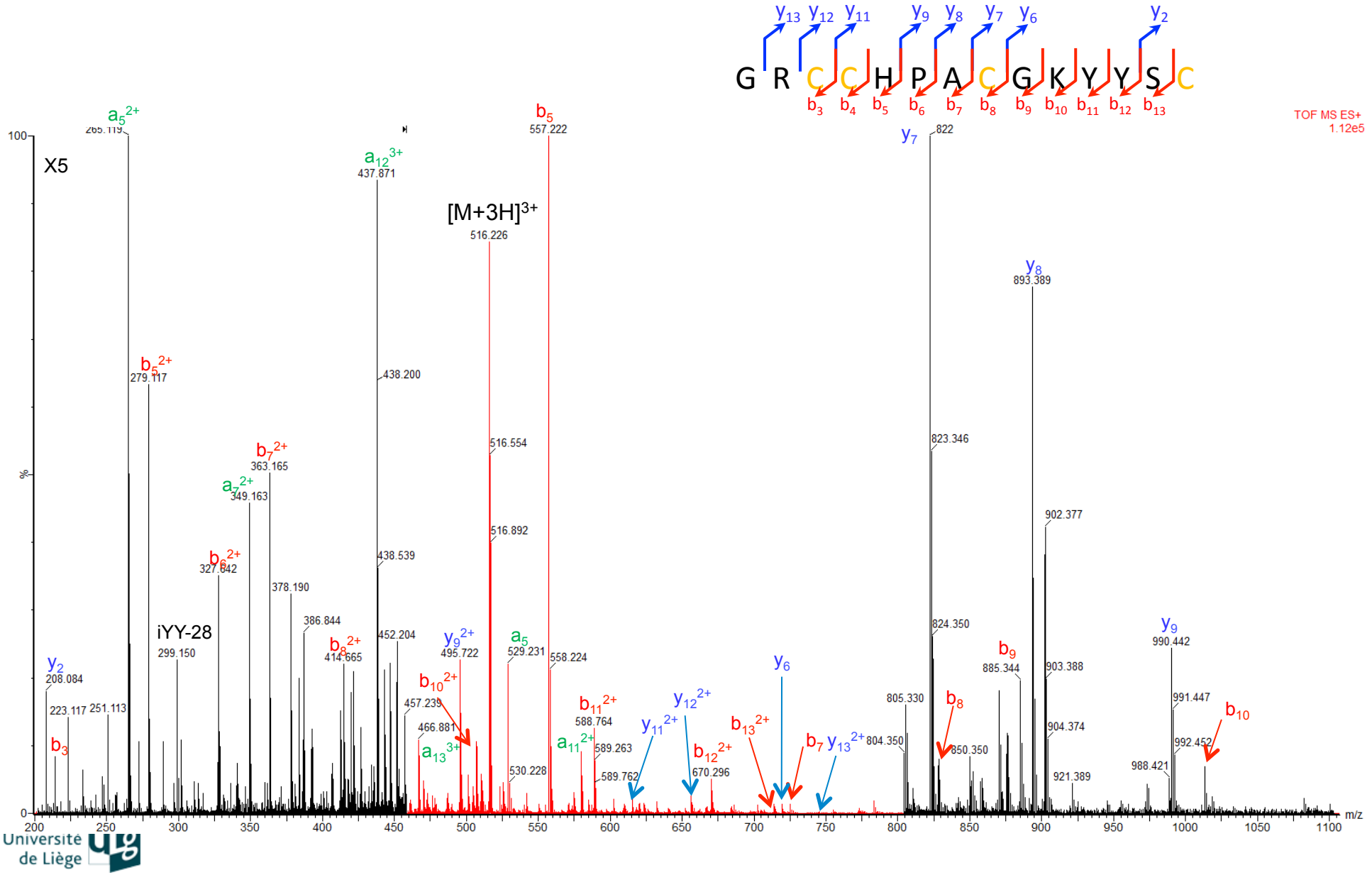
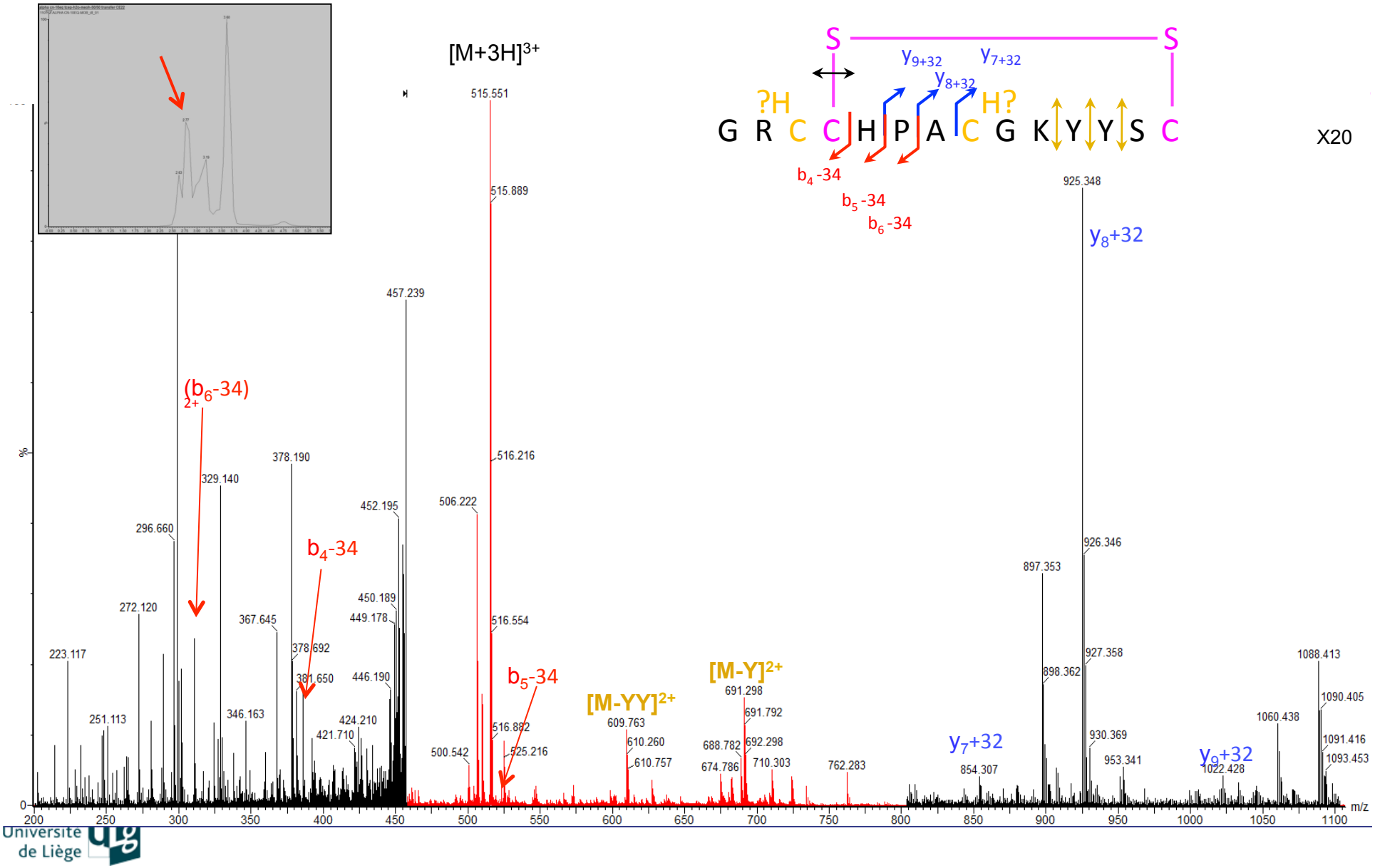
Fragmentation spectrum of the reduced form (arrival time : 3.532ms)



Fragmentation spectrum of the partially reduced form I (arrival time : 2.77ms)



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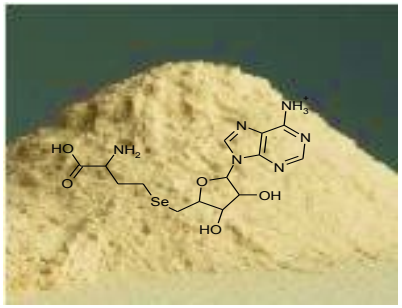
Fourth example

Selenium speciation

in enriched yeast



More details on poster n°20 Far *et al*.

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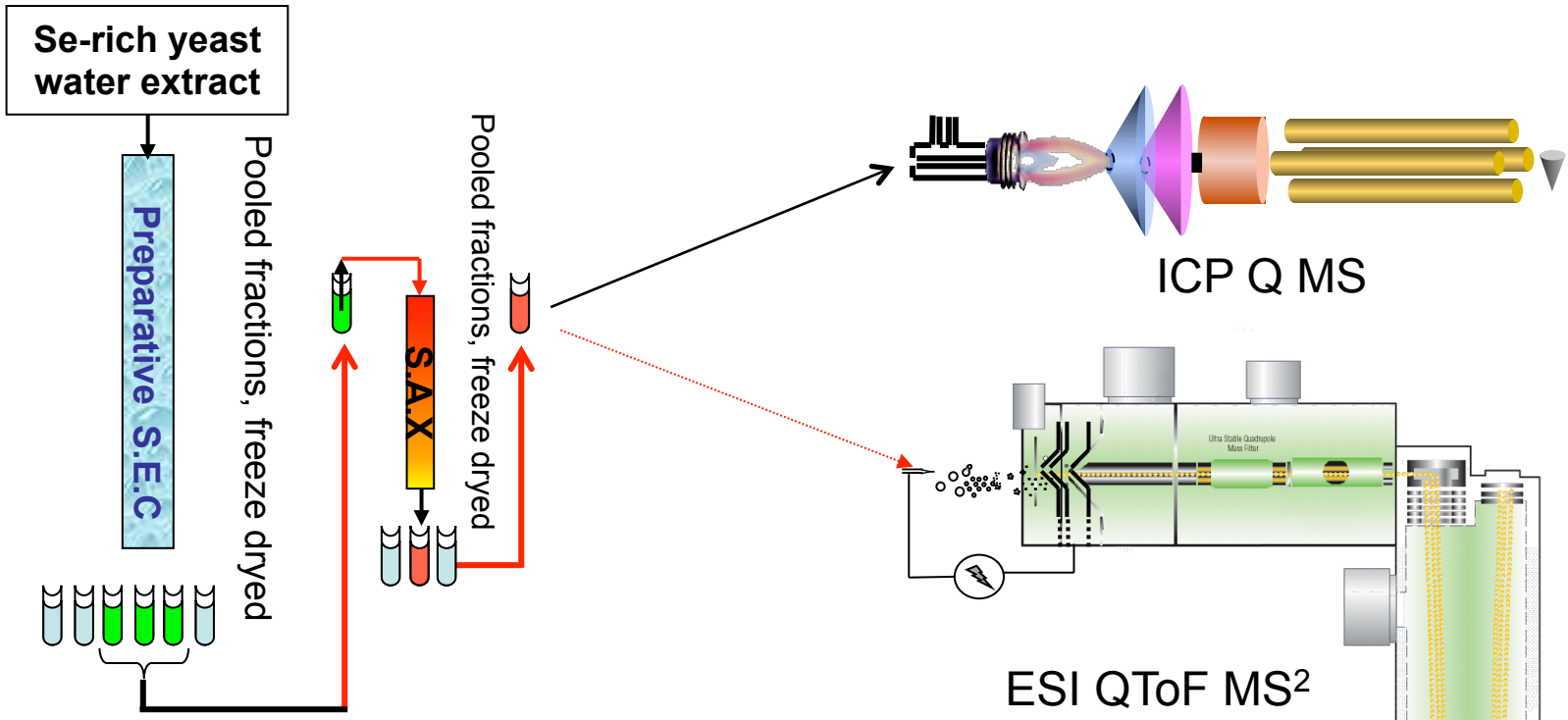
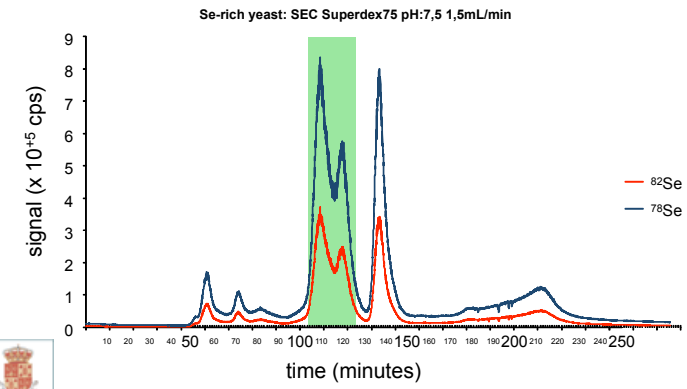
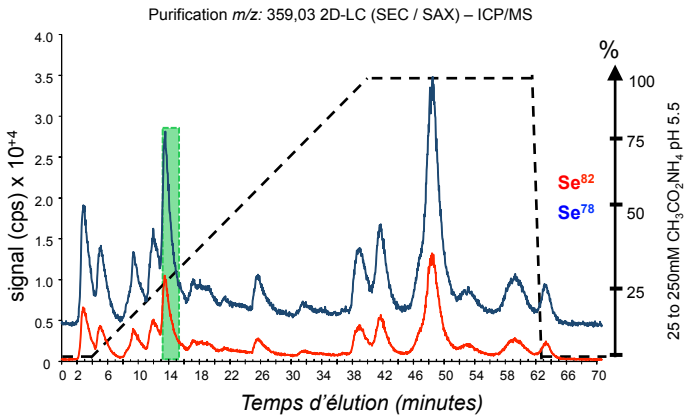
2 dimensions LC – MS approaches for Se-metabolites in yeast







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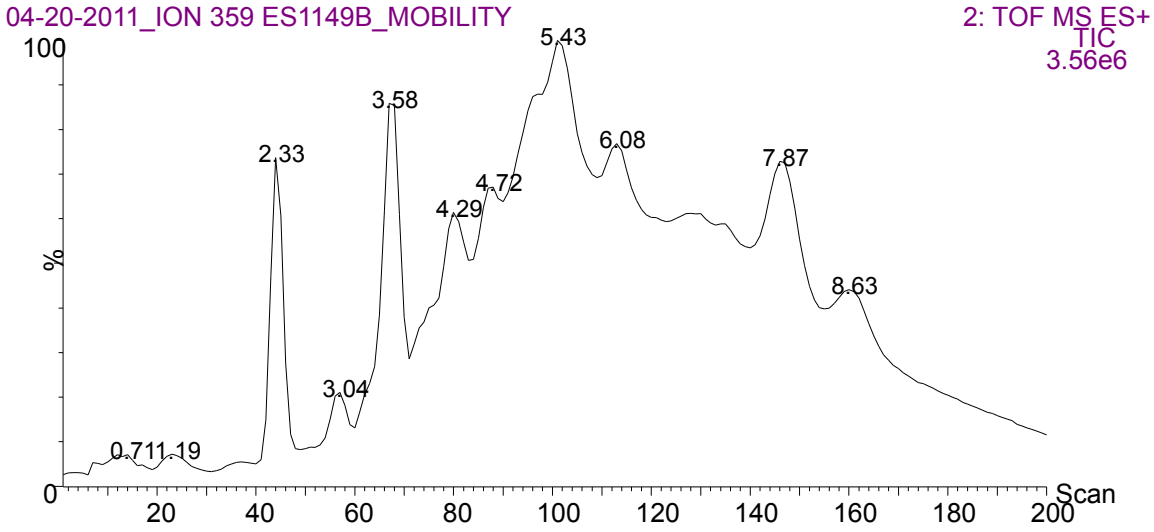
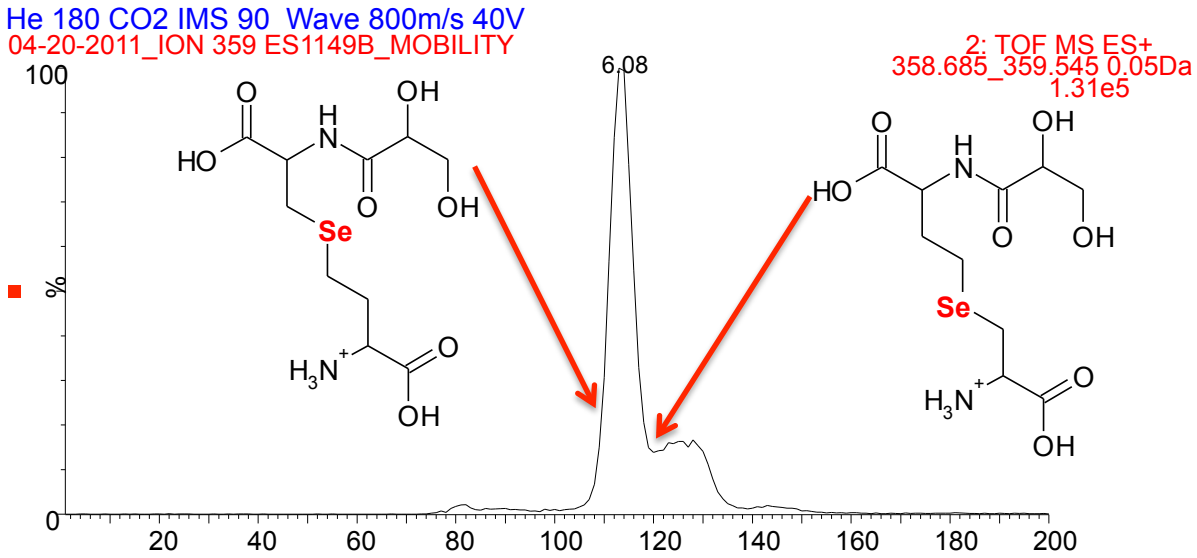
2D LC-ESI-IMS-MS





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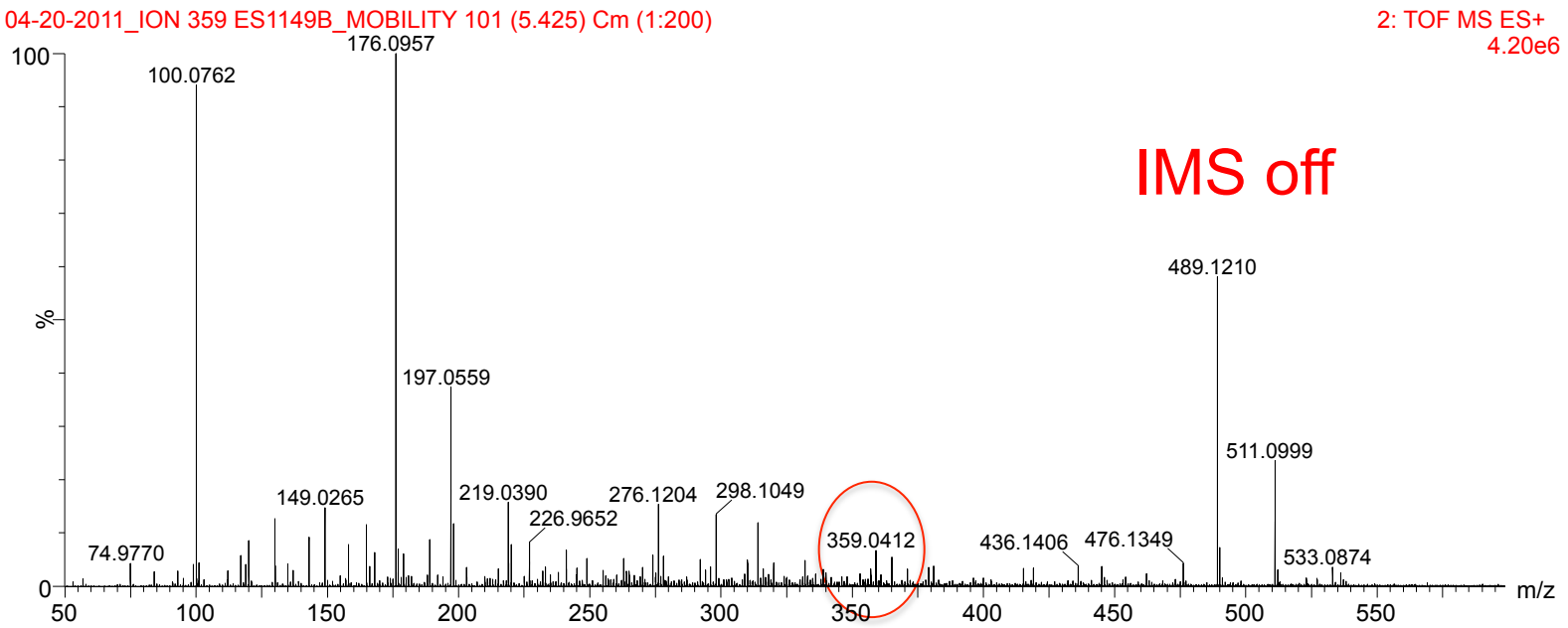

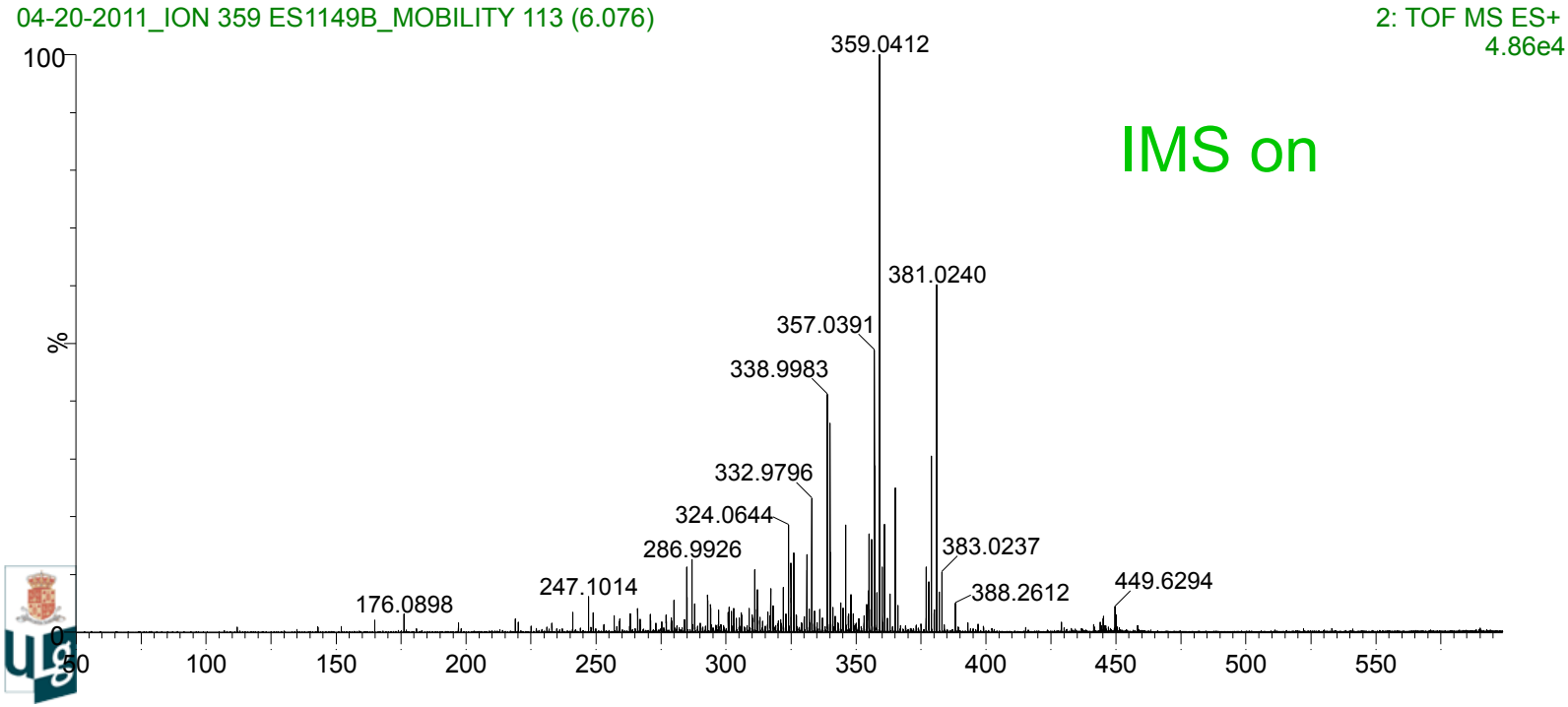

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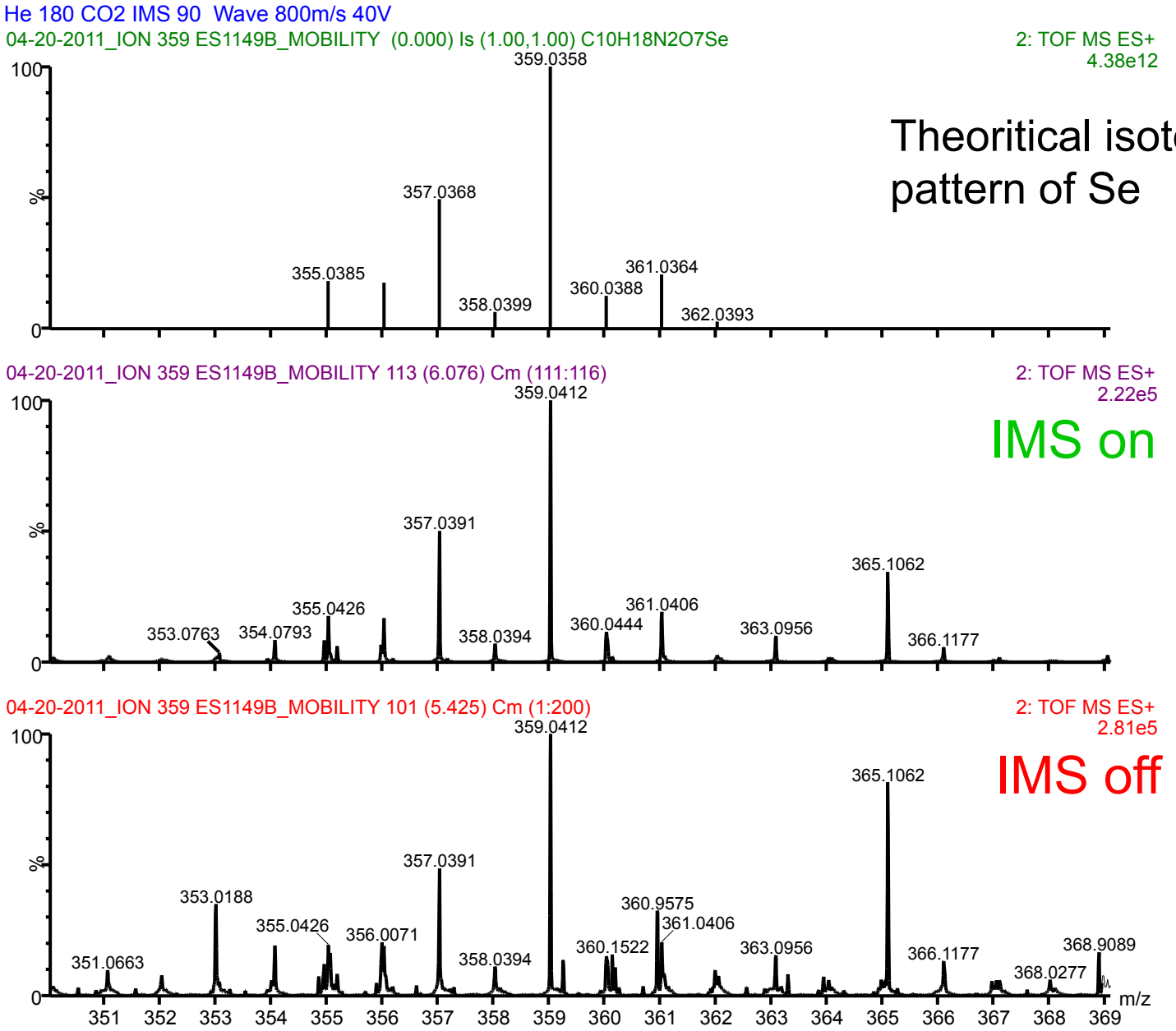
Isotope pattern of $C_{10}H_{18}N_2O_7Se + H^+$



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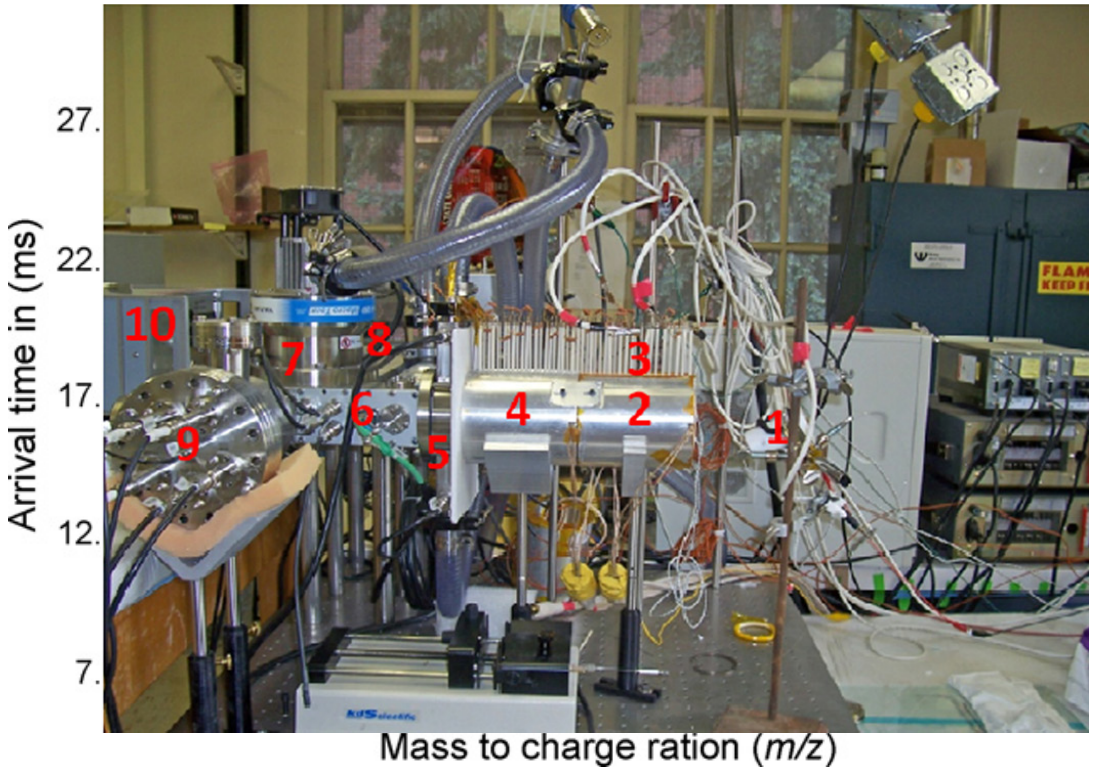
Application in metabolomics

Metabolic profiling of human blood plasma by high-resolution ion mobility mass spectrometry

Dwivedi et al. International Journal of Mass Spectrometry, 298, 2010, 78-90



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Molecular imaging

- The concept
- Small molecules
- Large molecules
- In situ identification

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MSI of Small molecules/Large molecules

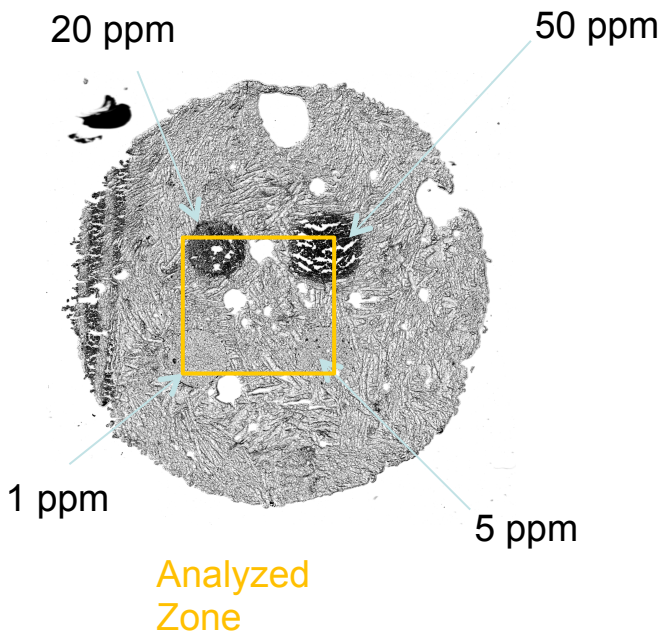
The requirements for imaging small molecules or large molecules differ:

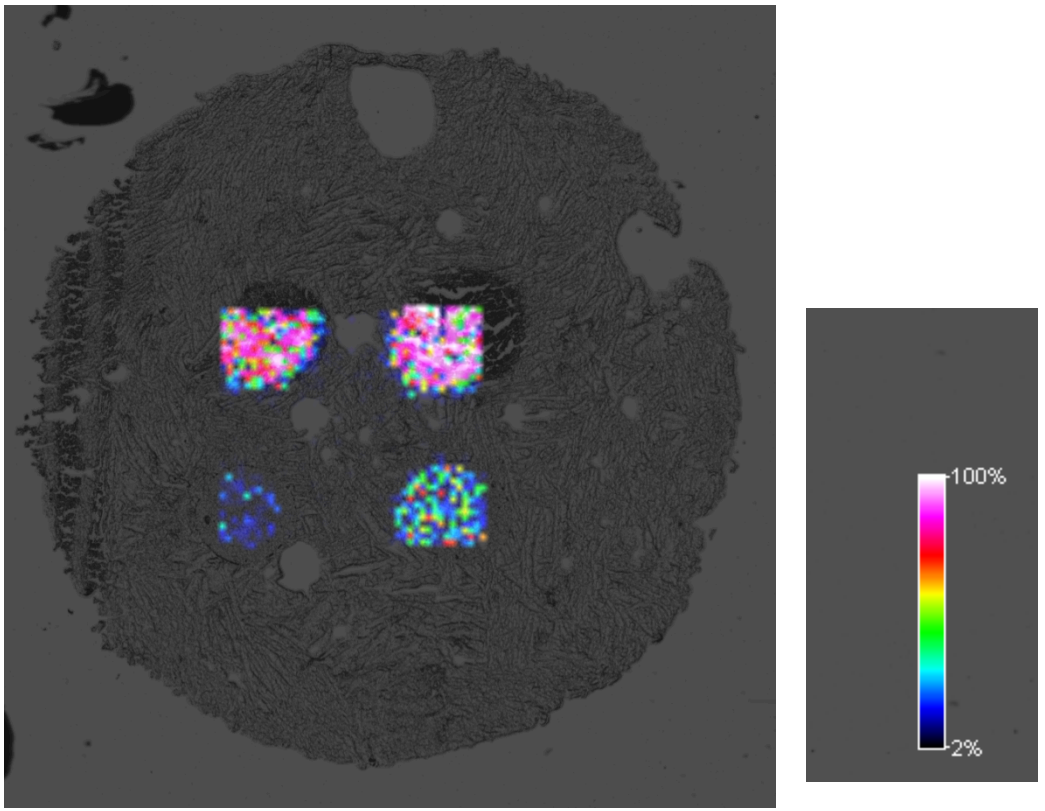
imaging: small molecules detection is made difficult in the presence of matrix interferences, large signal suppression due to the variety of chemicals, loss of spatial resolution in case of solubility in matrix solvents. Large molecules imaging is mostly dealing with peptides and proteins giving a good response.

identification: the identification of small molecules is straightforward at high resolution/accuracy, difficult for large molecules

Imaging of small molecules by MALDI -MS

Instrument: Bruker Daltonics Solarix
MALDI Source: Laser Smartbeam II 1000Hz, 500 shoots/pixel, 25% power, 100 µm raster
Sample: Pork Liver sausage (4 sampling zones spiked with 4 drugs: erlotinib, sunitinib (anticancer), reserpine et terfenadine (antihistaminic drug) at different levels of concentration





Imaging of sunitinib (m/z 399.2191)

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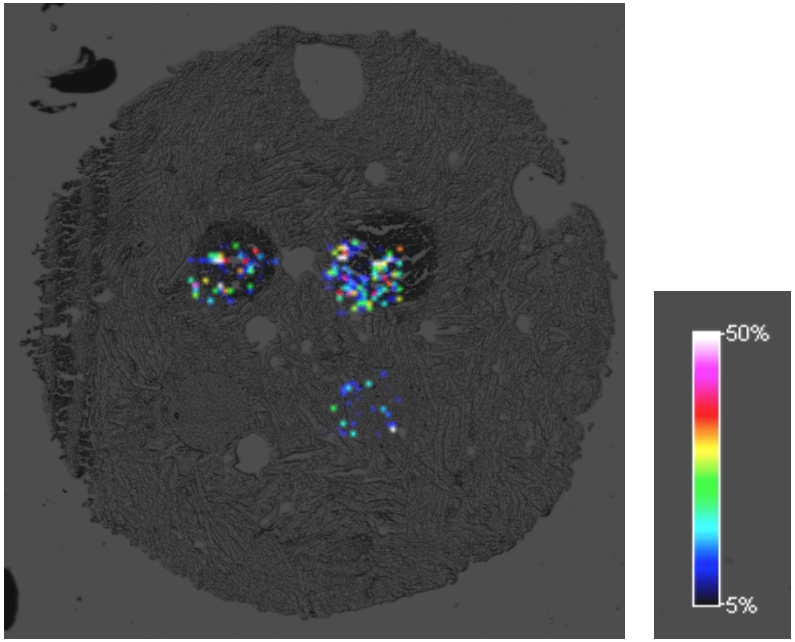
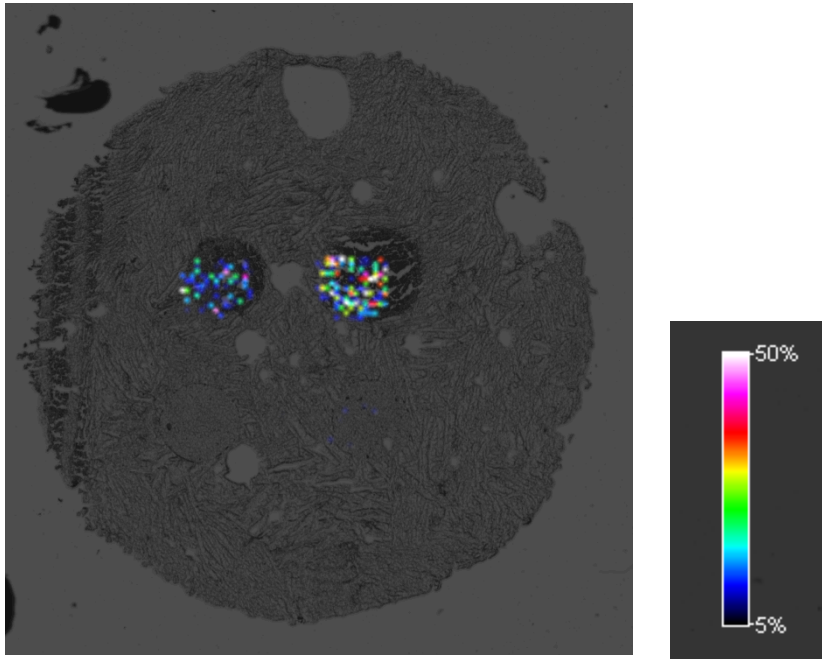


Imaging of erlotinib (m/z 394.1765)



Imaging of reserpine (m/z 609.2820)

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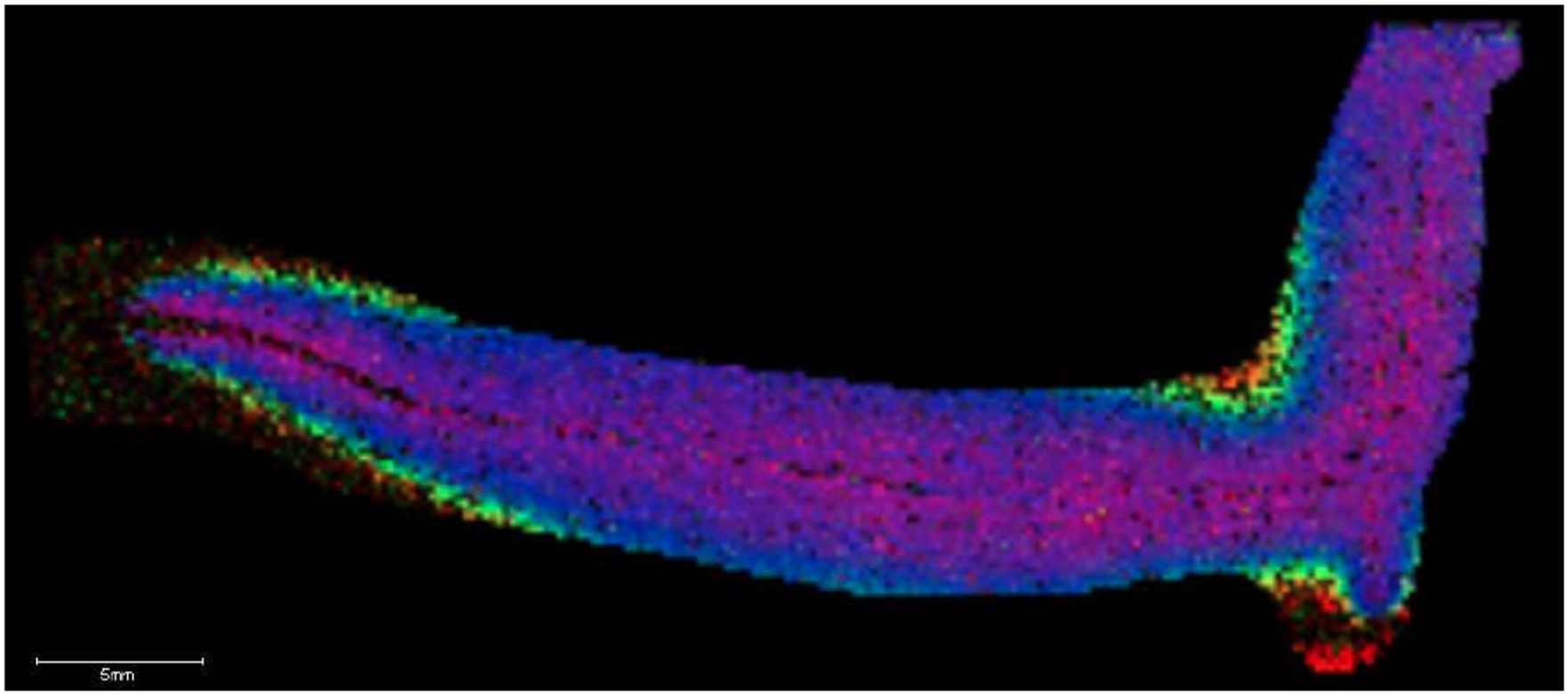
MALDI MS imaging of tomato root



Superposition of pictures of 4 surfactins (lipopeptide)
Surfactin C_{12} in red on edge (far from the root)
Surfactin C_{13} in green
Surfactin C_{14} in blue, very abundant
Surfactin C_{15} in red, in the neighborhood of the root

Thanks to

- Edwin De Pauw (Director of LSM)
- Romain Touilloux/Laure Joly/Séverine Goscinny (Pesticides)
- Julie Echeterbille/Loic Quinton (Venomics)
- Gabriel Mazzucchelli (MS)
- Frédéric Rosu (MM calculations)
- Johann Far (Selenometabolites)

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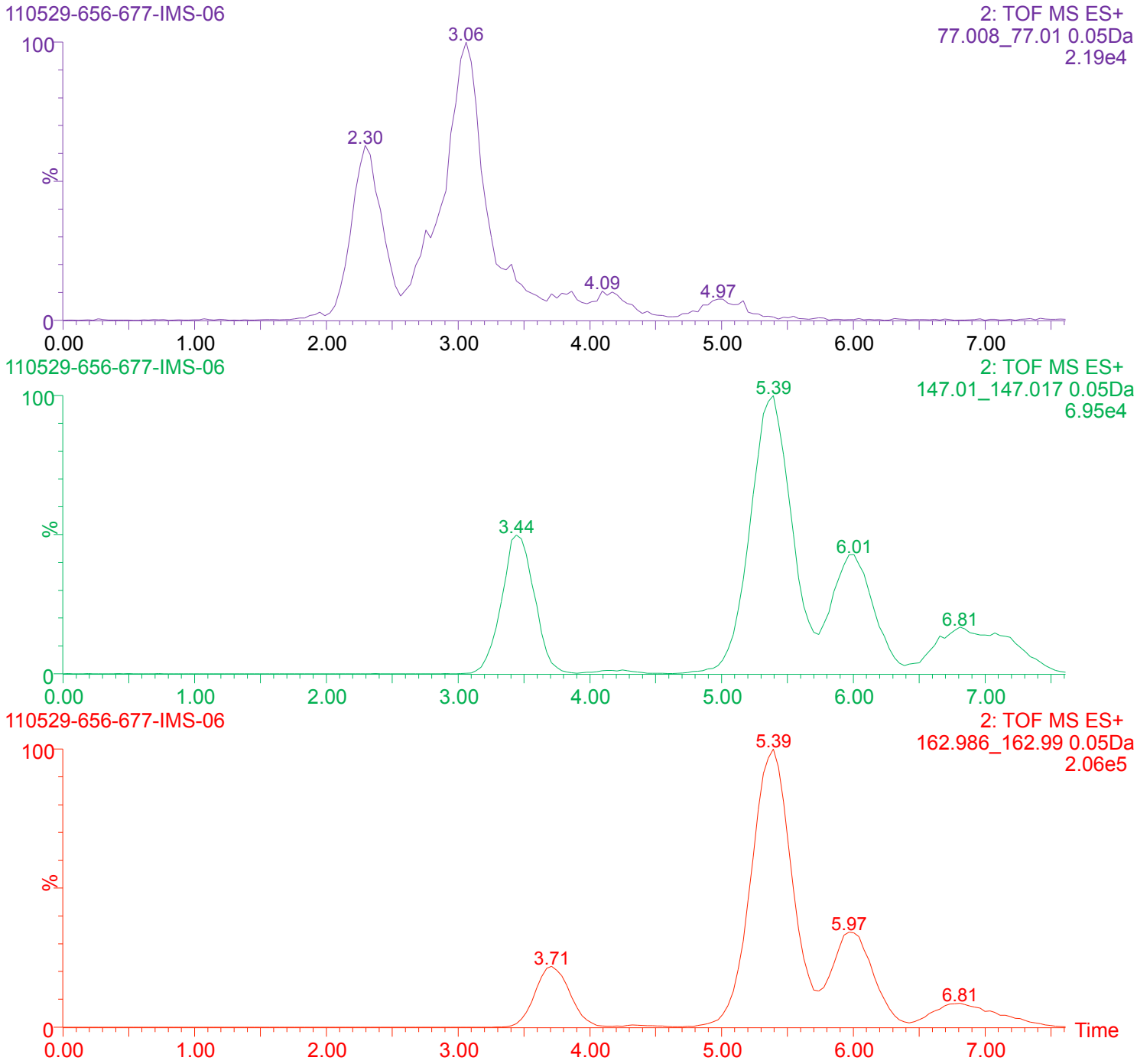

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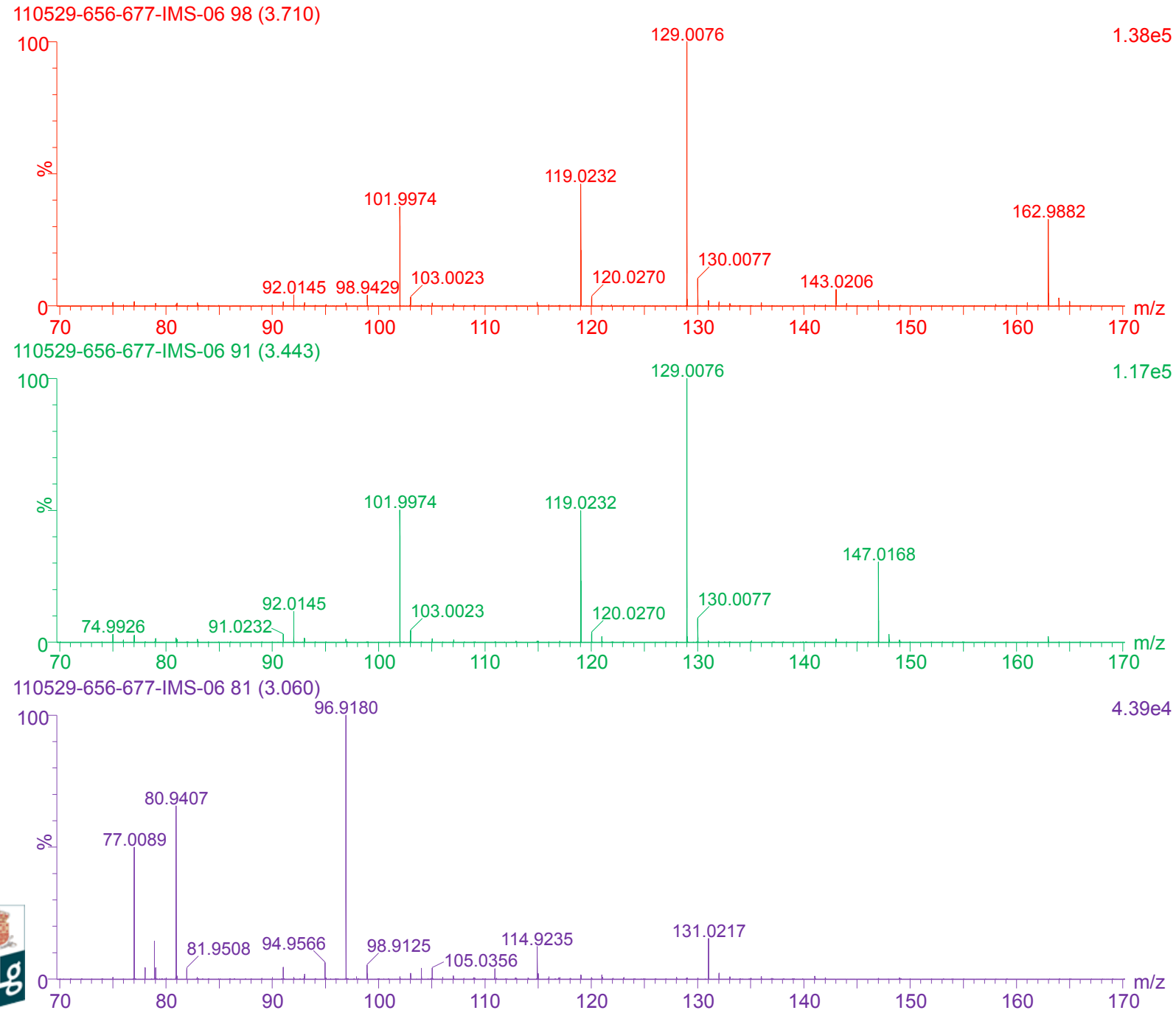

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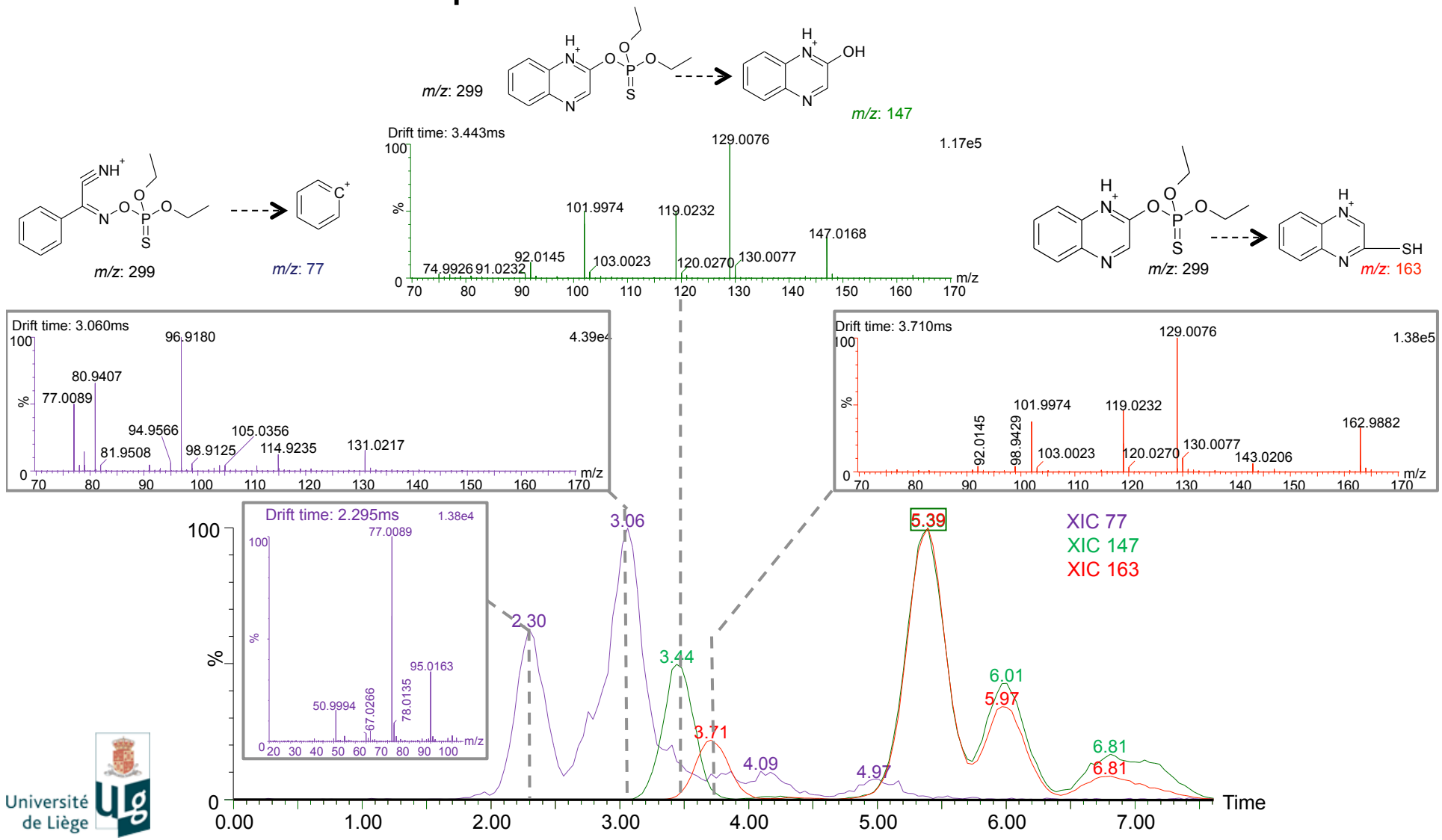
ESI – Trap MS/MS – IMS – Transfer MS/MS



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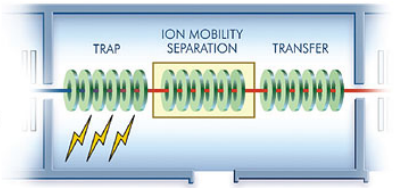
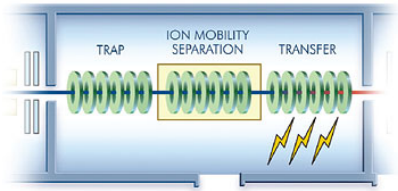
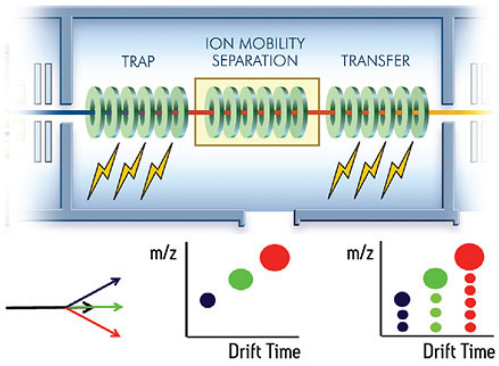
TRAP
T-Wave
fragmentation





TRANSFER
T-Wave
fragmentation

TAP fragmentation



1) Precursor ion fragmented

2) Product ions separated by IMS

3) 1st and 2nd generation products are **Time Aligned**

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TWIGS ok but…

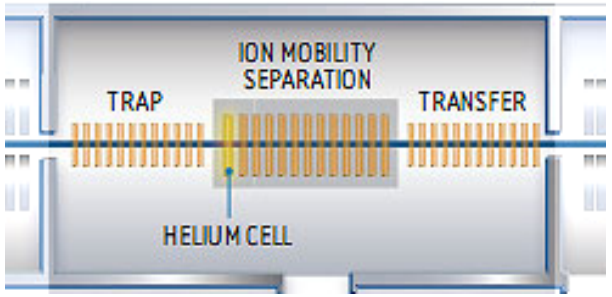
1. Ions can be vibrationally excited with an increase higher than 200K in the effective temperature; the activation can take place at different places where voltages are applied or within the IMS, according to the wave height

2. This can induce fragmentation but could also changes ions conformation . A high bias could induce changes trapped upon cooling in the IMS

To minimize this effect:

Within the IM:

- Reduction of the wave height
- Increase of the wave velocity
- Increase of the gas pressure



In front of the IM

- Use of a light gas curtain

3. As mobility is related to the temperature, this may induce errors in measurements or, once controlled, be used as structural tool, the IMS being considered as a reactor