Swedish multiresidue method SweEt goes into Orbitrap technology

Susanne Ekroth

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”Nothing is more simple than greatness; indeed, to be simple is to be great.”

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Outline

- Introduction, Uppsala and NFA
- Objectives
- Comparisons to LC-MS/MS and TOF
- Examples and Results
- Future plans
- Conclusions, Pros and Cons

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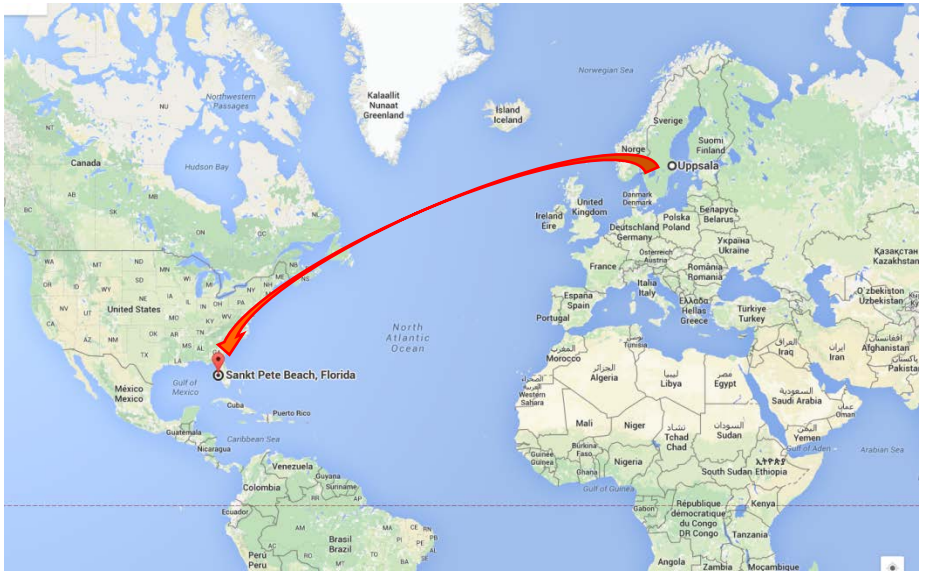
Uppsala



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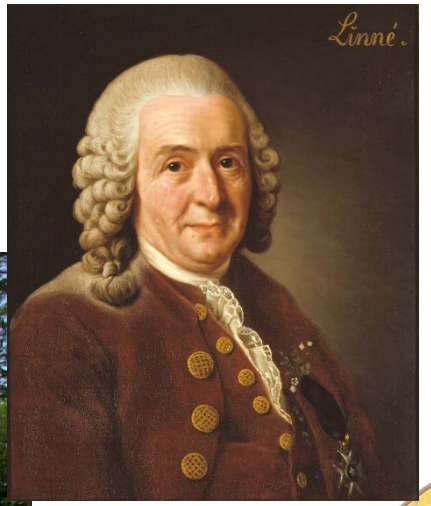
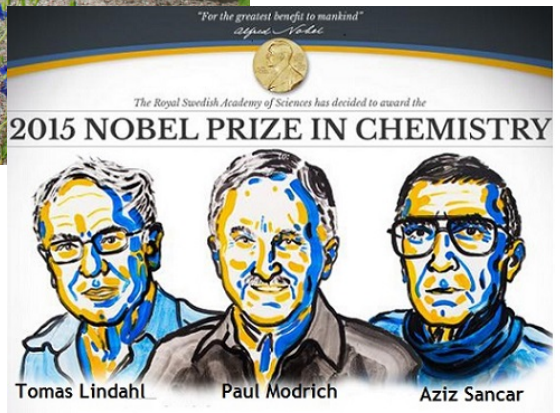
Carl von Linné





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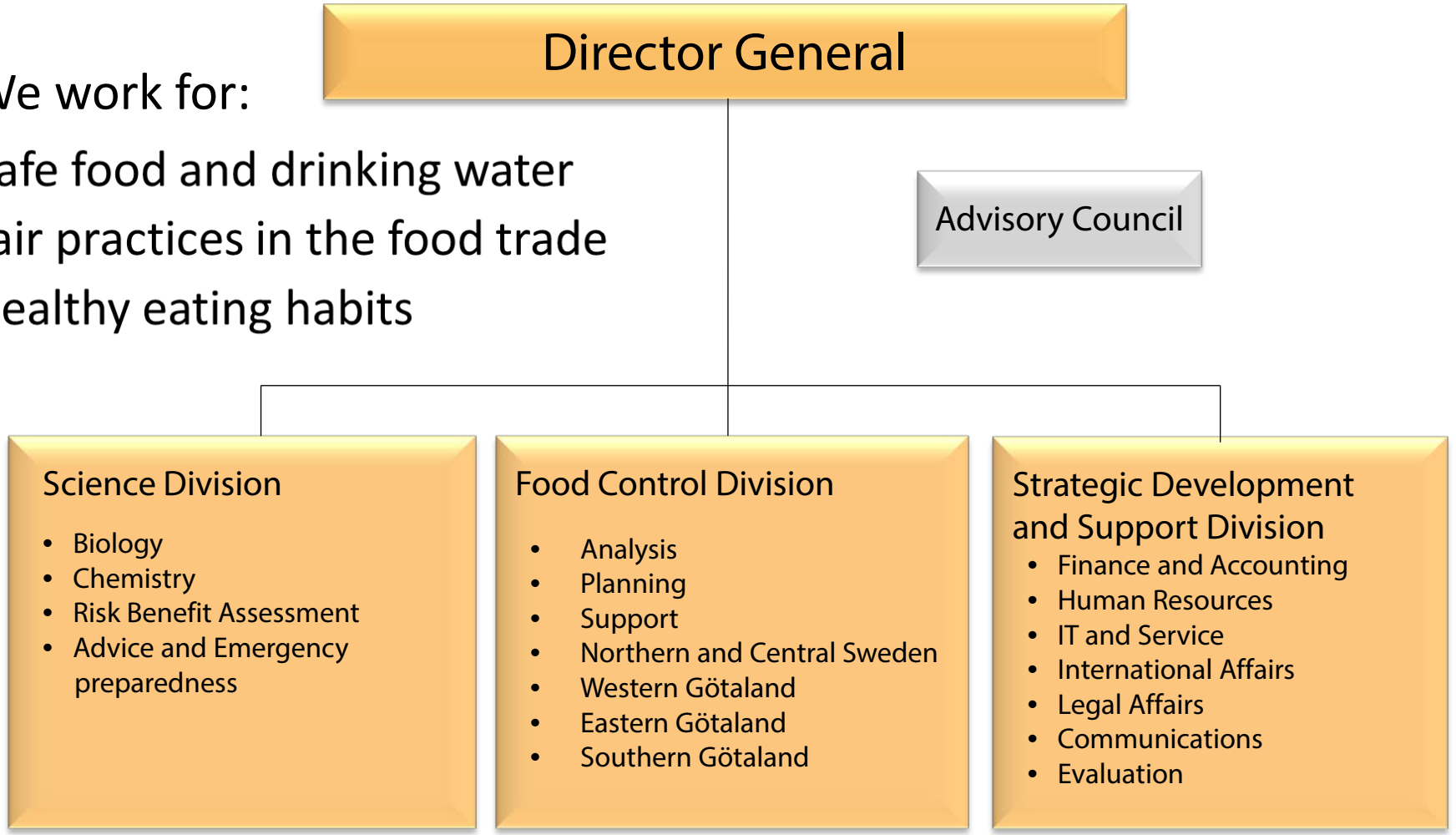
The National Food Agency

We work for:

- safe food and drinking water
- fair practices in the food trade
- healthy eating habits



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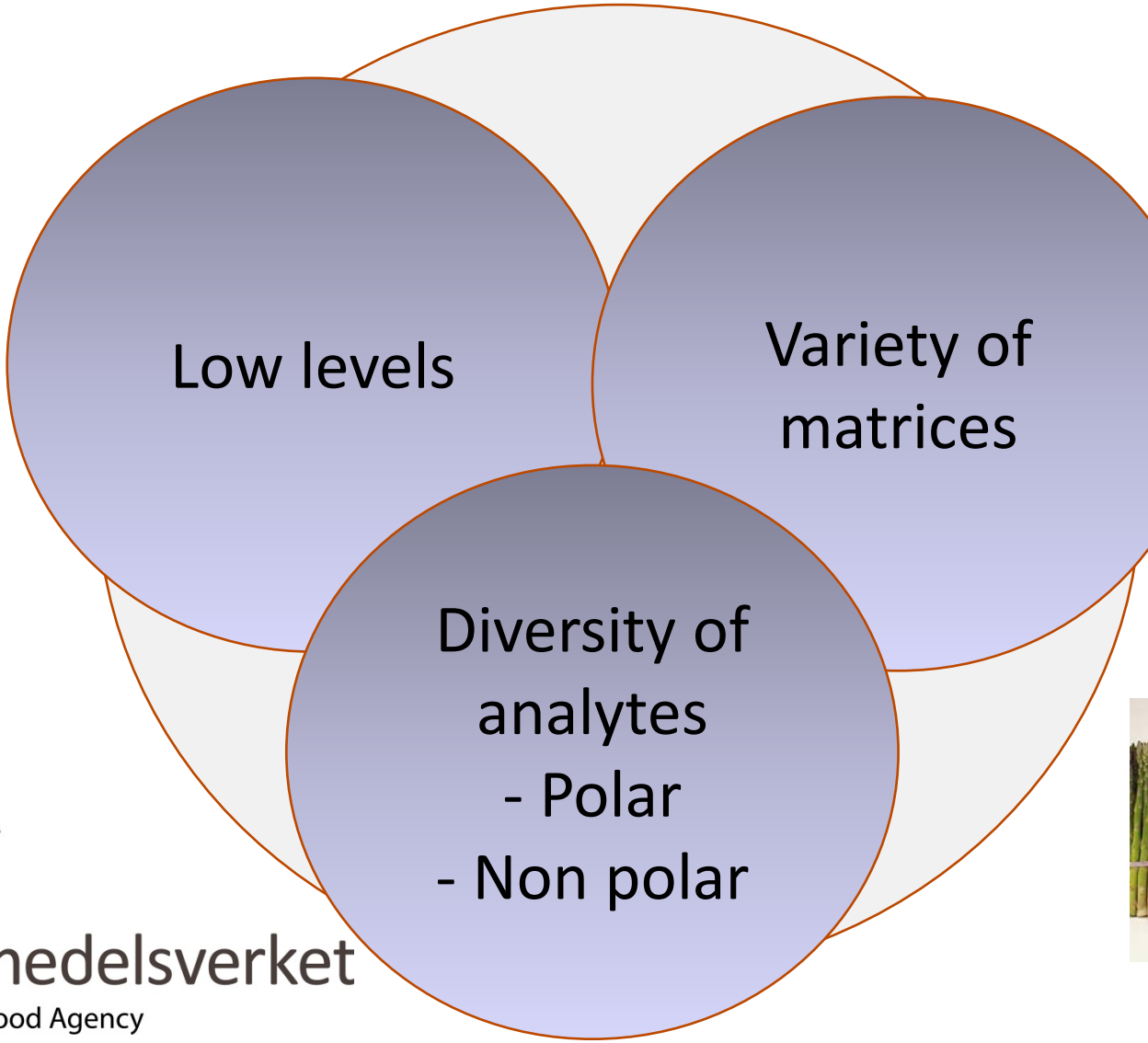
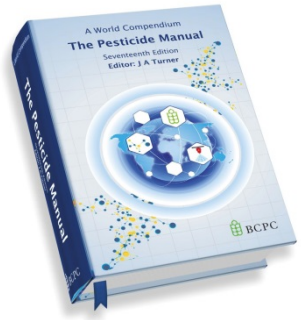
Challenges working with pesticides





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Validation and Control procedures for Pesticide laboratories

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SweEt

FV+Honey

448

Extraction

10 g sample
20 ml EtOAc+$NaHCO_3$
3 min Spex shaker
Na_2SO_4

Centrifugation

Filtration

GC-MS/MS

UPLC-MS/MS + HRMS

Cereals

323

Extraction

5 g sample
10 ml water
10 ml EtOAc (1% HAc)
3 min Spex shaker
Na_2SO_4

Centrifugation

Filtration

GC-MS/MS

UPLC-MS/MS

AO<10%fat

96

Extraction

5 g sample
PSA + C18
10 ml EtOAc
3 min Spex shaker
Na_2SO_4

Centrifugation

Filtration

GC-MS/MS

UPLC-MS/MS

AO>10% fat

76

Fat determination

Extraction

0.5 g fat
10 ml EtOAc:CH (1+1)
3 min Spex shaker
Na_2SO_4

Centrifugation

Filtration

GPC SX-3
(1+1 EtOAc/CH)

Evaporation

Reconstitution in EtOAc

GC-MS/MS

UPLC-MS/MS

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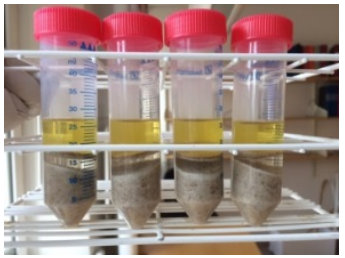
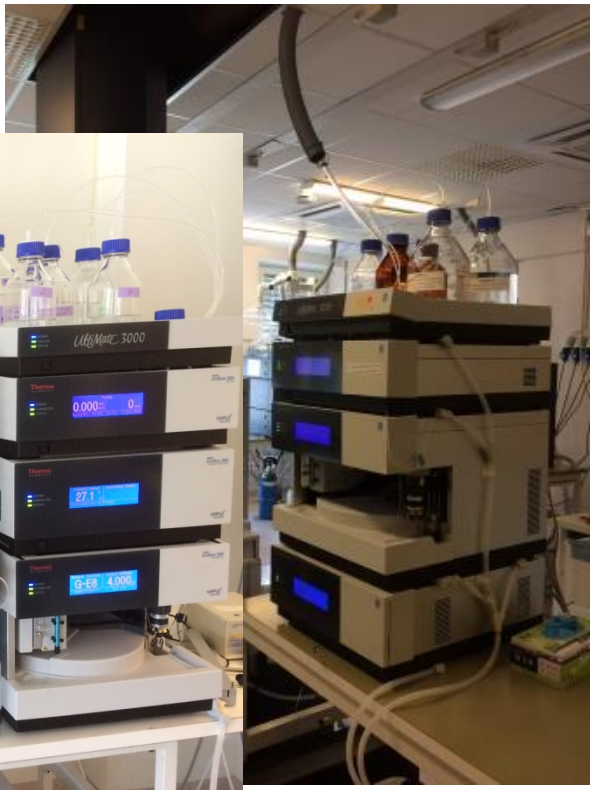
SweEt - Workflow





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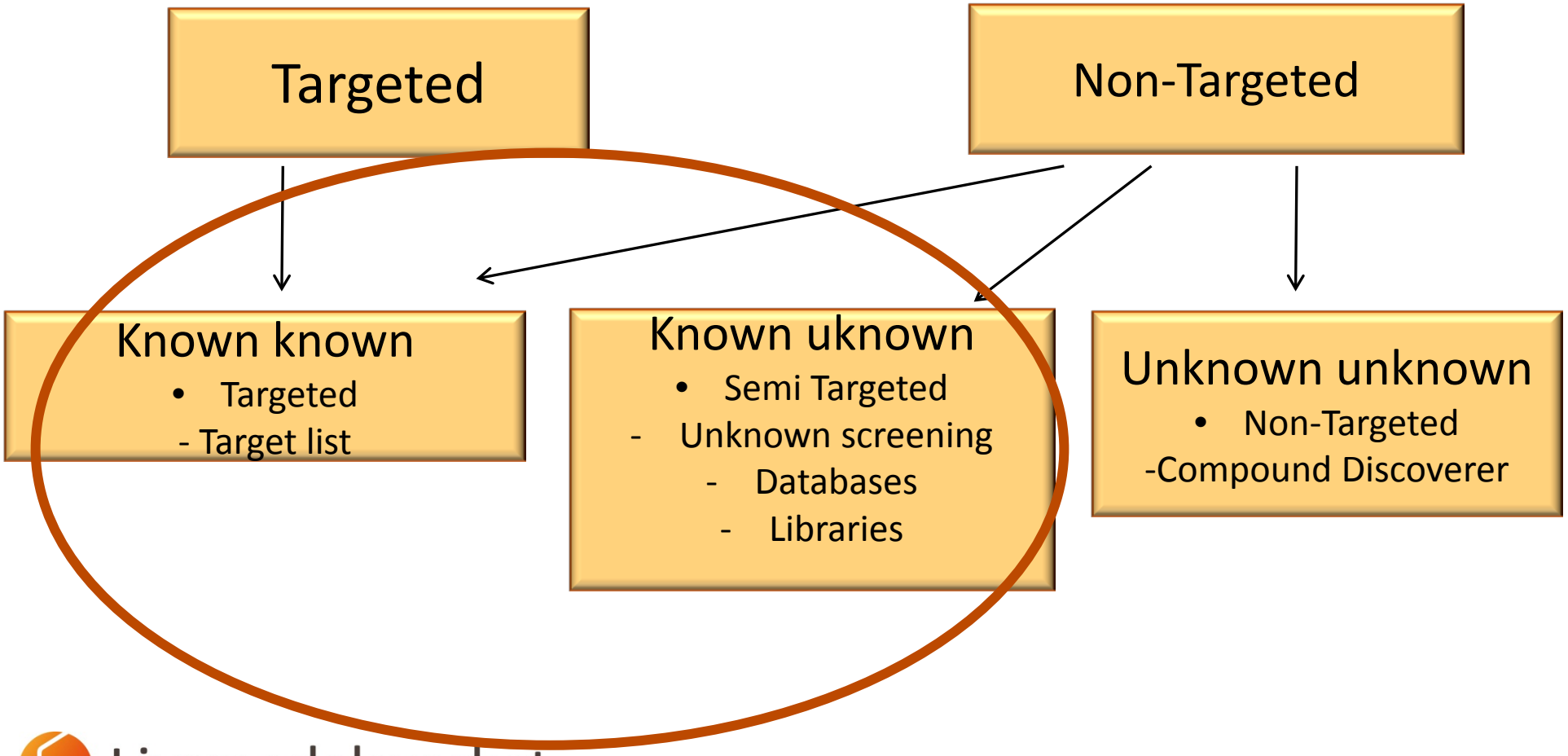
Workflow for Targeted and Non-Targeted Analyses



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Verification of Q Exactive Focus Orbitrap

- ☑ Linearity
- ☑ Robustness
- ☑ Detectability

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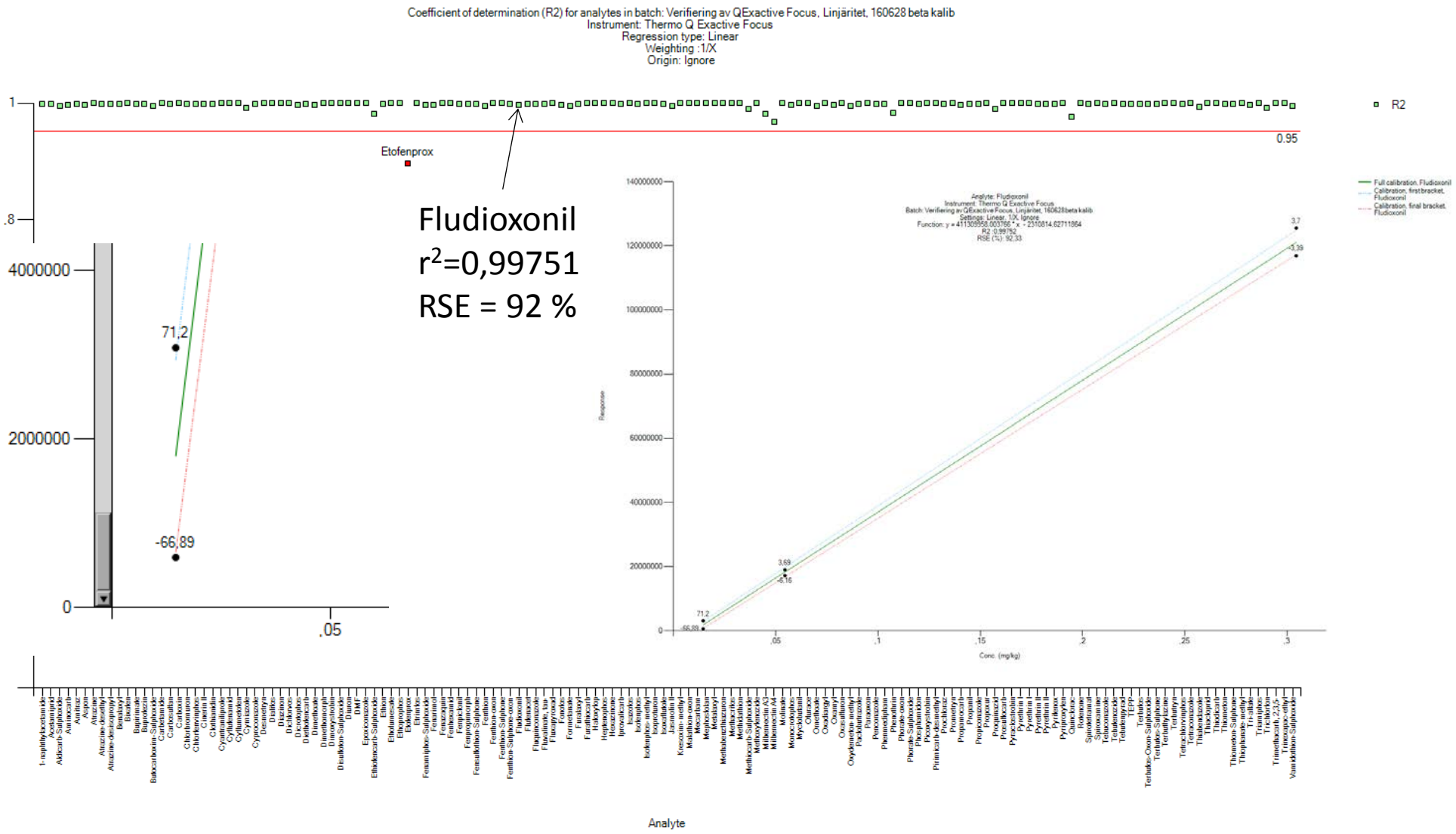
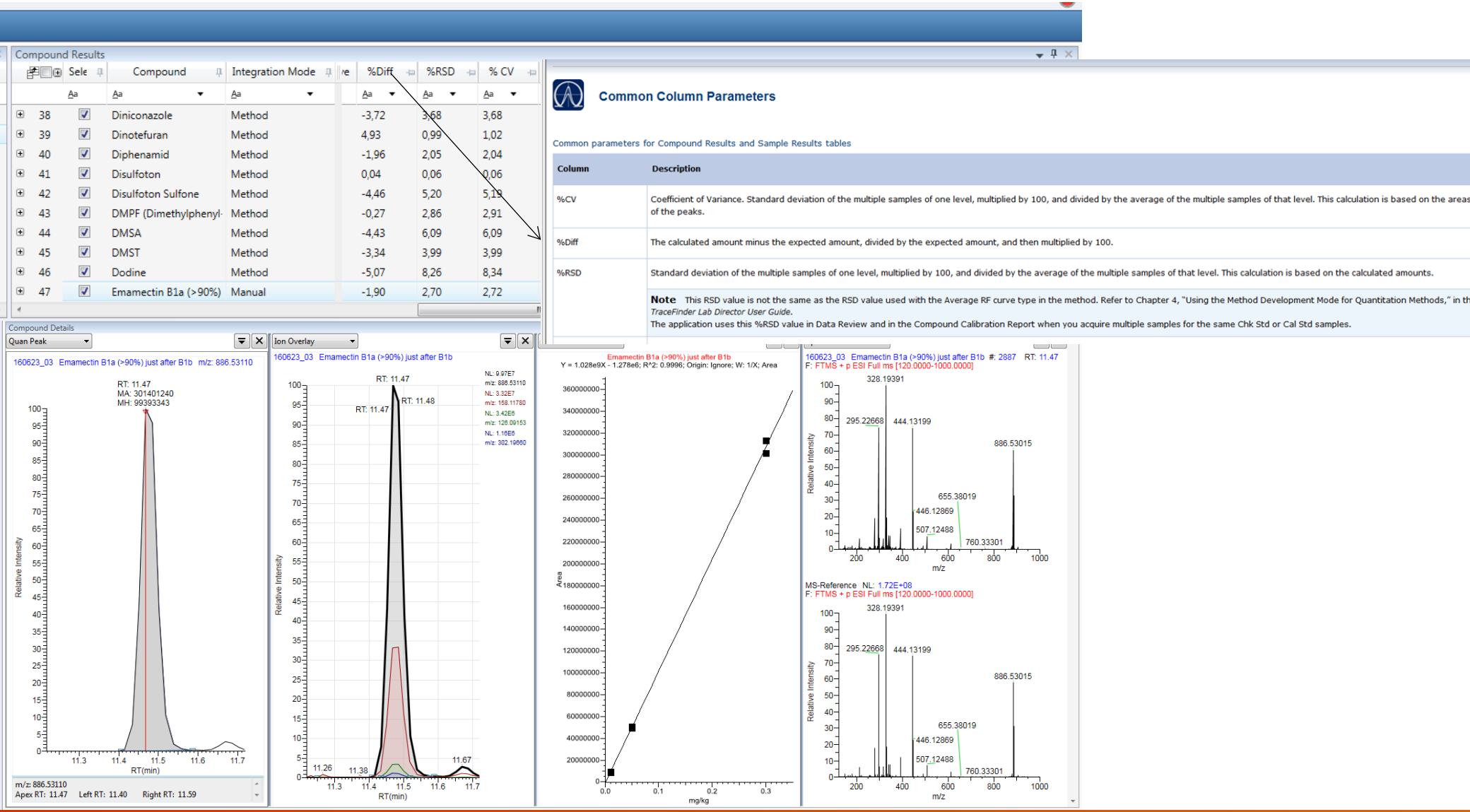
Linearity – correlation or residuals?



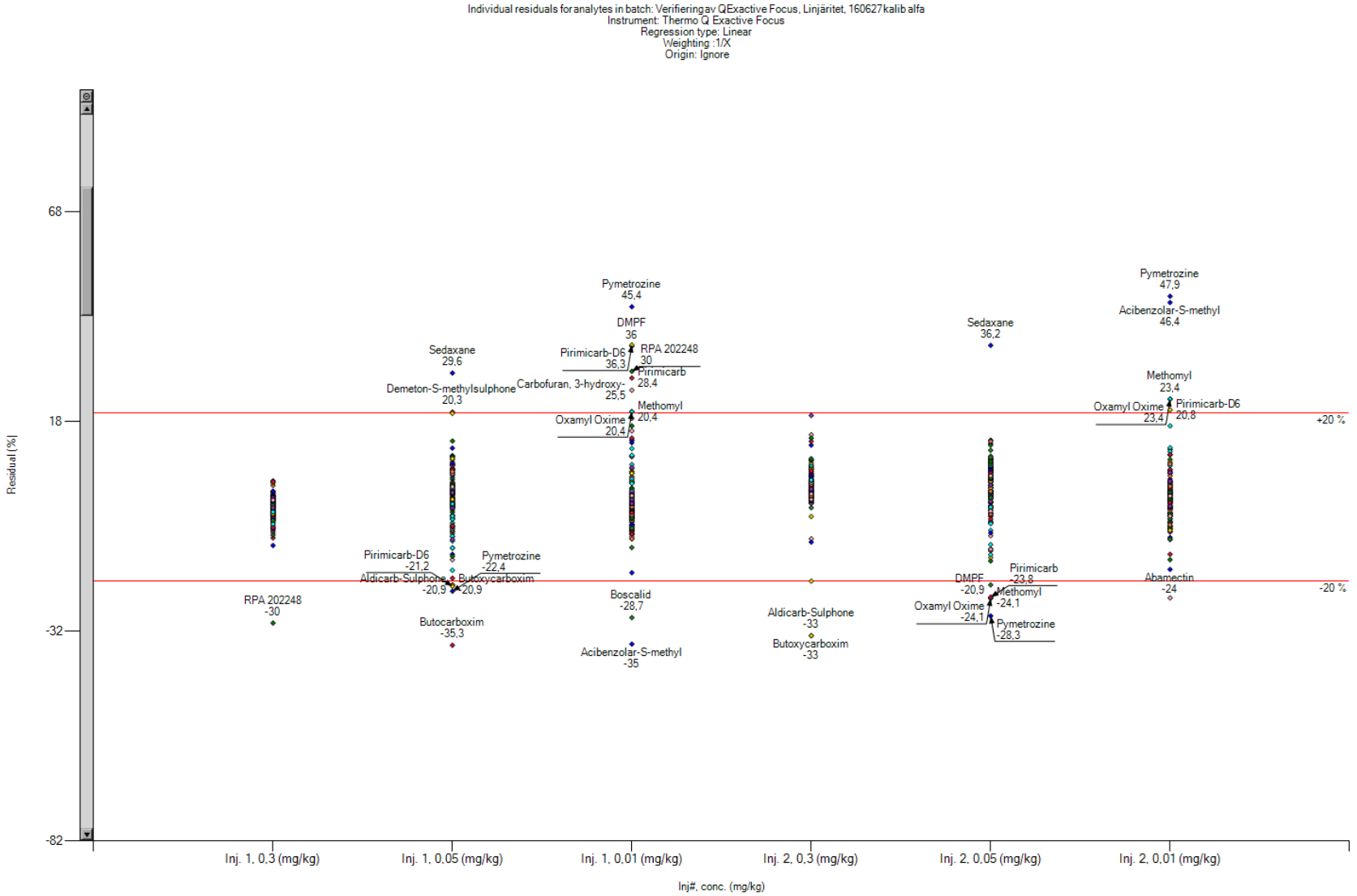
Linearity according to SANTE doc

C17 Multi-level calibration (three or more concentrations) is preferred. An appropriate calibration function must be used and the calibration curve should not be forced through the origin without justification. The fit of the calibration function must be plotted and inspected visually and/or by calculation of the residuals, avoiding over-reliance on correlation coefficients, to ensure that the fit is satisfactory within the concentration range of the pesticides detected. If individual residuals deviate by more than ±20% from the calibration curve in the relevant region, an alternative calibration function must be used. In general, the use of weighted linear regression (1/x) is recommended, rather than linear regression.

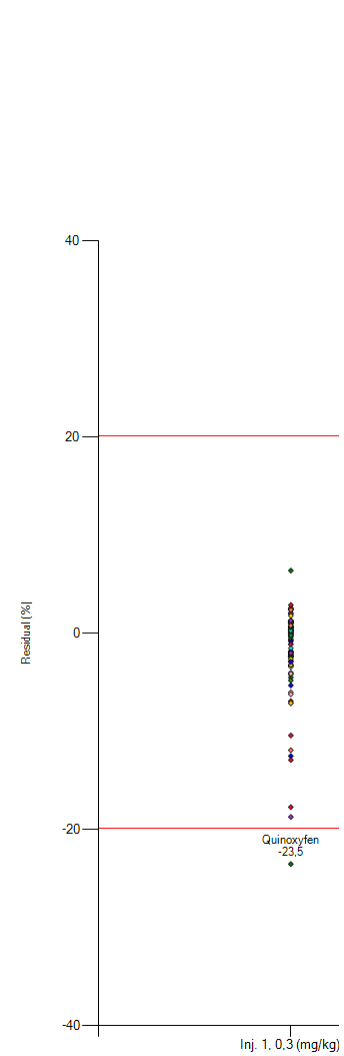
Quantitative View – with residuals

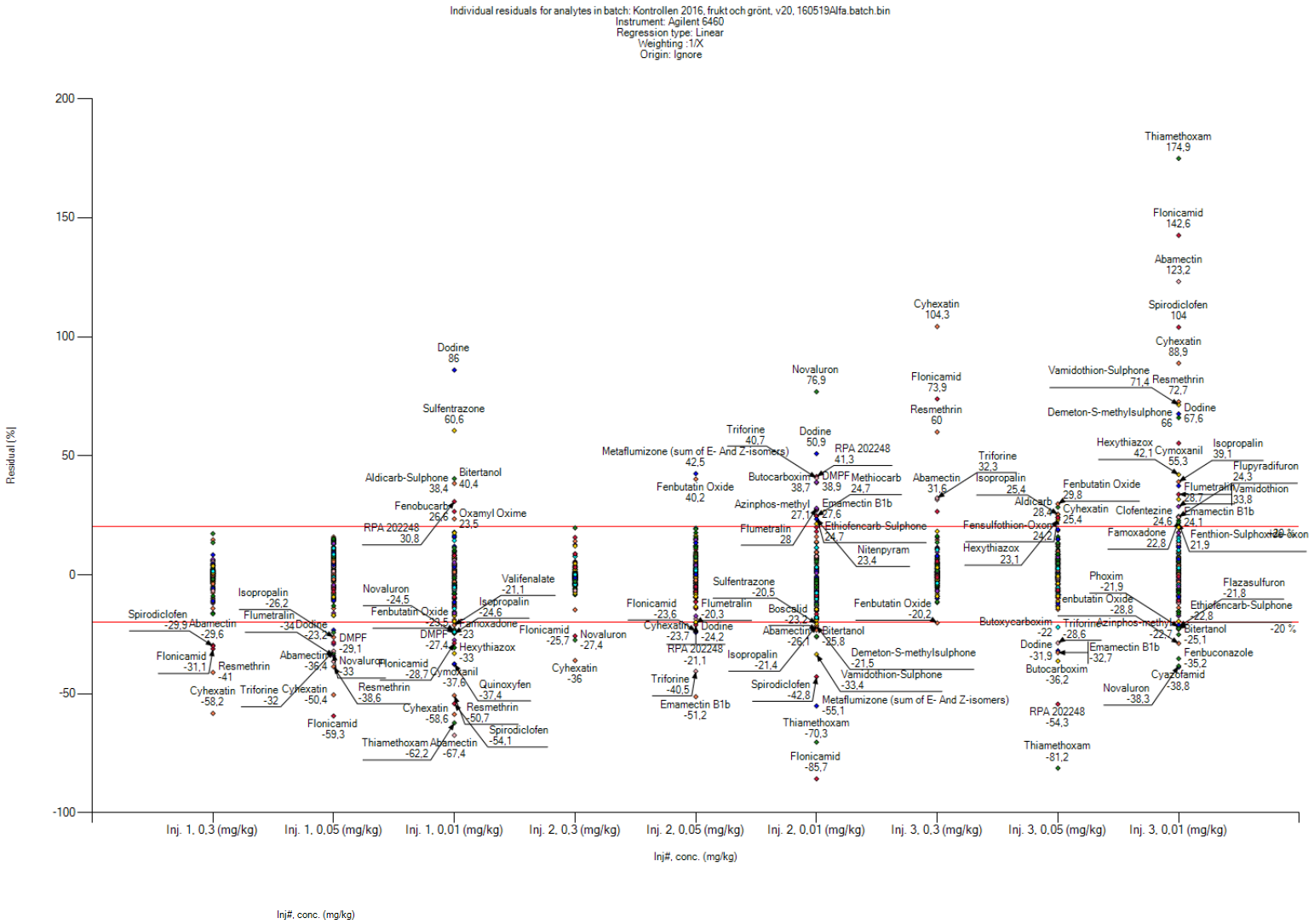


Residuals with Q Exactive Focus Orbitrap

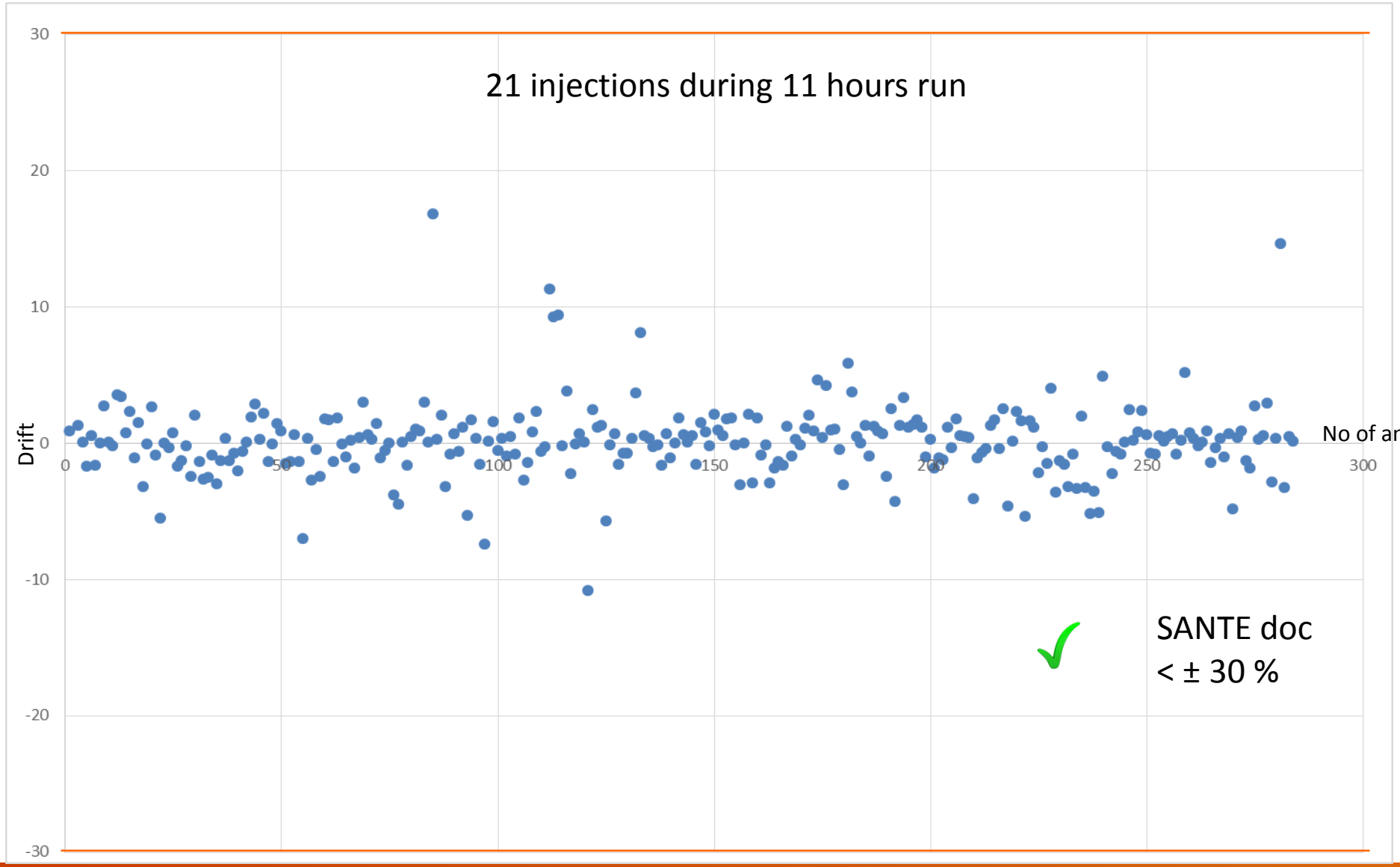


Residuals with current LC-MS/MS system





Robustness of the system

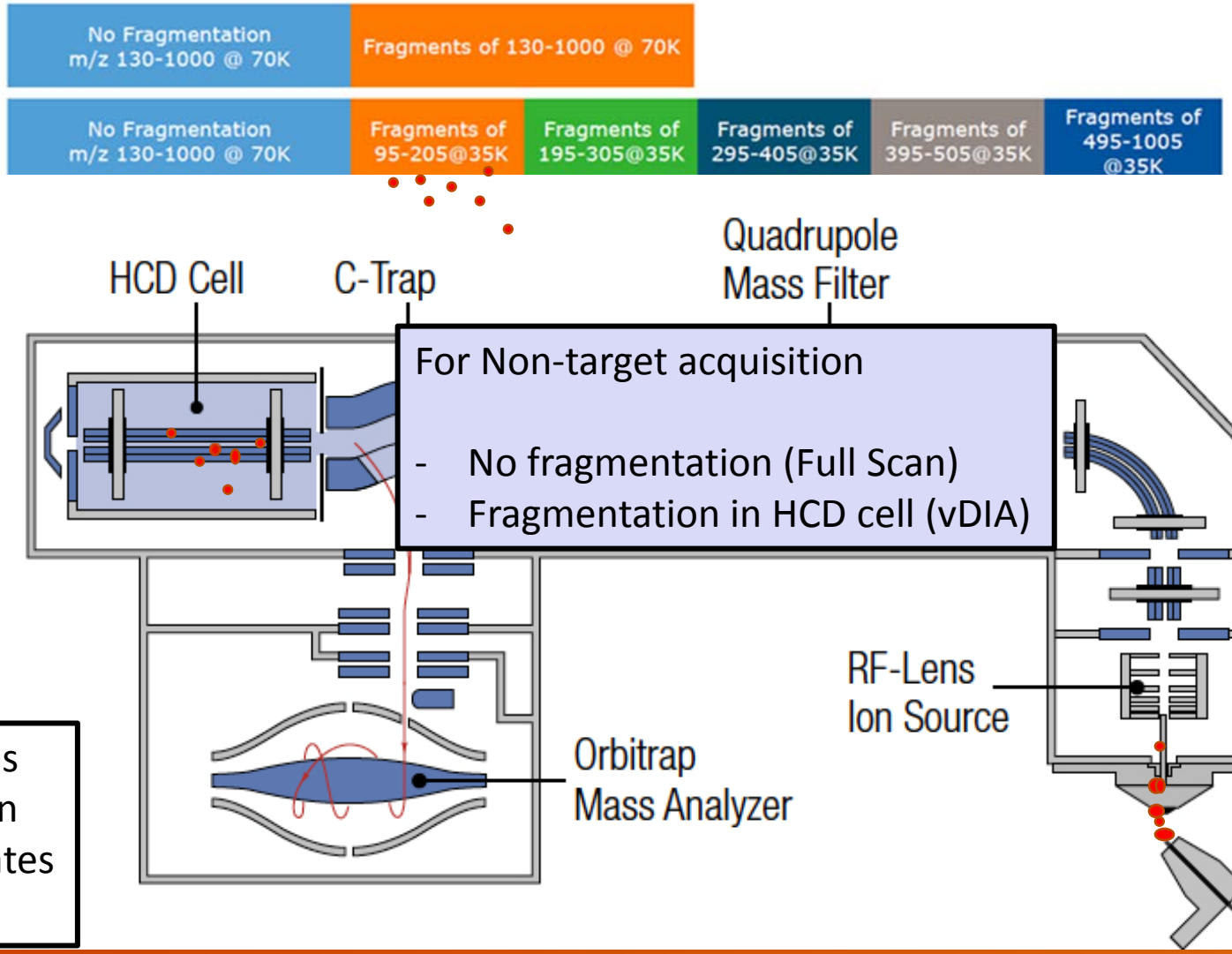


Detectability

EUPT-CF10 2016 wheat calculated with carrot

		LC-MS/MS	Orbitrap
Pesticide	**Assign value mg/kg**	**Reported amount Wheat calib mg/kg (z-score)**	**Calculated amount Carrot calib mg/kg (z-score)**
Azoxystrobin	0.088	0.073 (-0.7)	0.069 (-0.8)
Bixafen	0.074	0.059 (-0.8)	0.072 (-0.1)
Buprofezin	0.048	0.051 (0.2)	0.052 (-0.3)
Tebuconazole	0.091	0.080 (-0.5)	0.076 (-0.6)
Epoxiconazole	0.177	0.174 (-0.1)	0.154 (-0.5)
Pirimicarb-desmethyl	0.048	0.055 (0.6)	0.047 (-0.1)
Pyraclostrobin	0.098	0.081 (-0.7)	0.077 (-0.8)
Tetramethrin	0.096	0.102 (0.3)	0.094(-0.1)
Isocarbophos	0.048	0.057 (0.7)	0.042 (0.6)
Fluopyram	0.250	0.269 (0.3)	0.223 (-0.4)
Prothioconazole-desthio	0.149	0.149 (0)	0.130 (0.6)

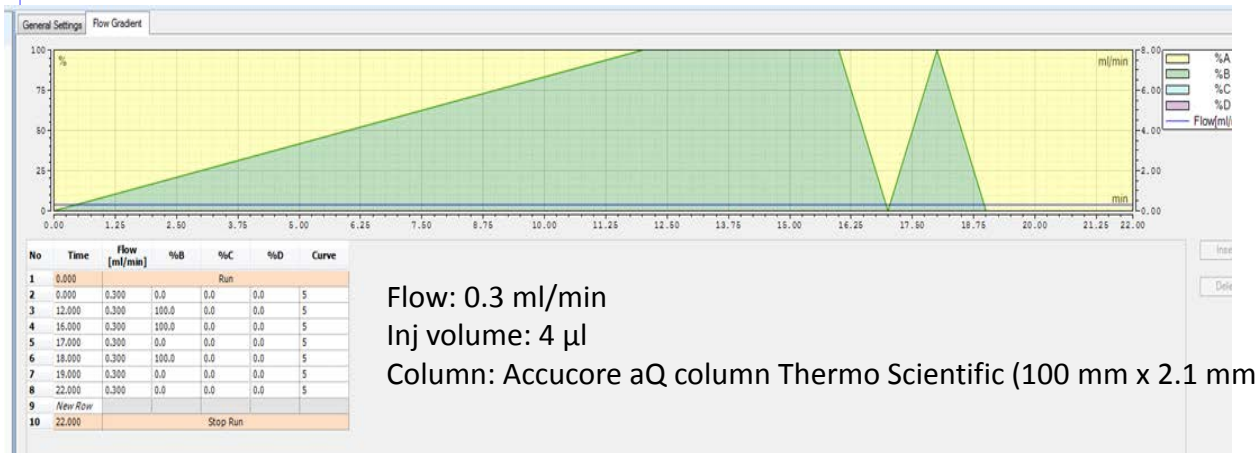
Full scan combined with vDIA acquisition



vDIA method is not available in the United States of America

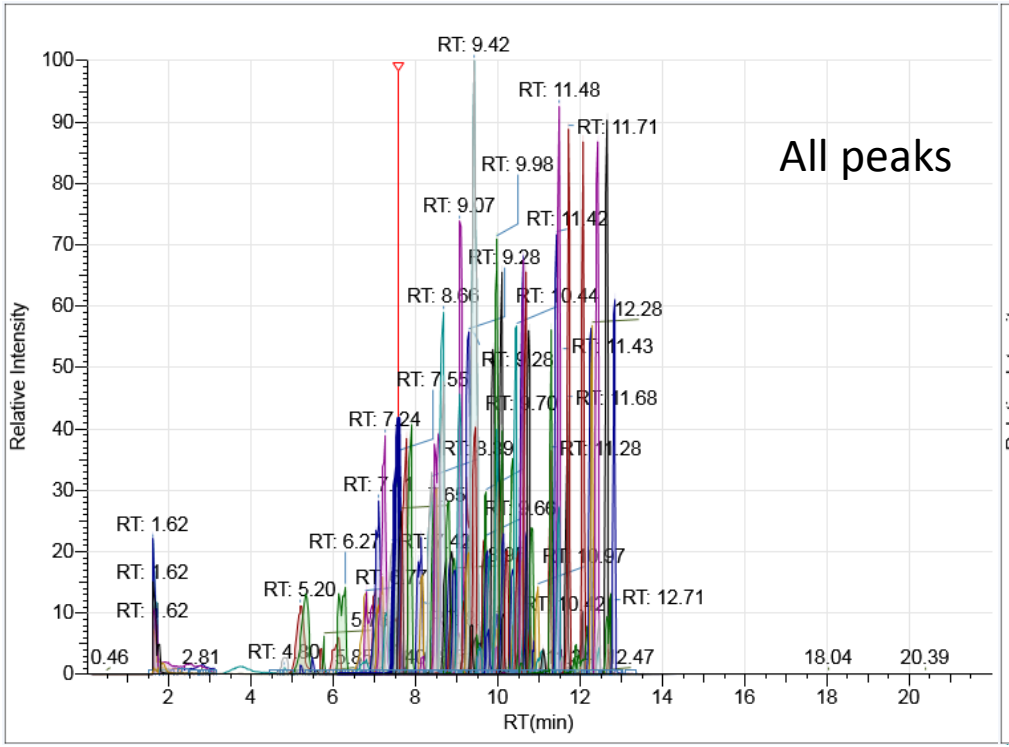
Properties

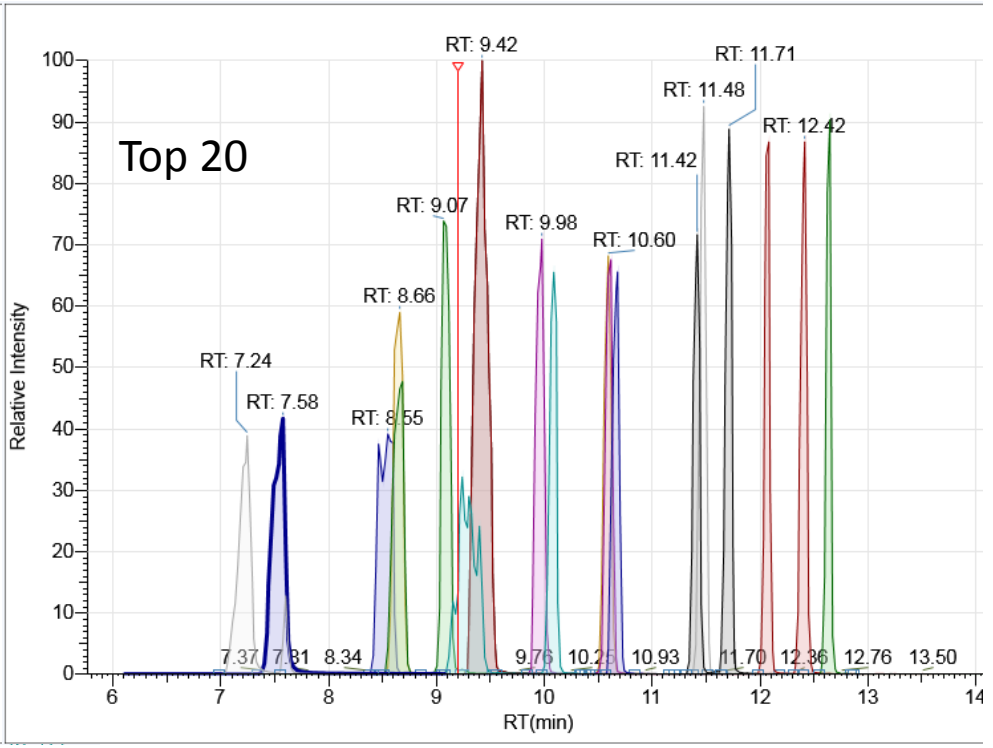
Mobile phase:
A: 5mM ammonium formiate and 0.1 % formic acid in water
B: 5 mM ammonium formiate and 0.1 % formic acid in methanol



Flow: 0.3 ml/min
Inj volume: 4 µl
Column: Accucore aQ column Thermo Scientific (100 mm x 2.1 mm ID)

XIC Overlay





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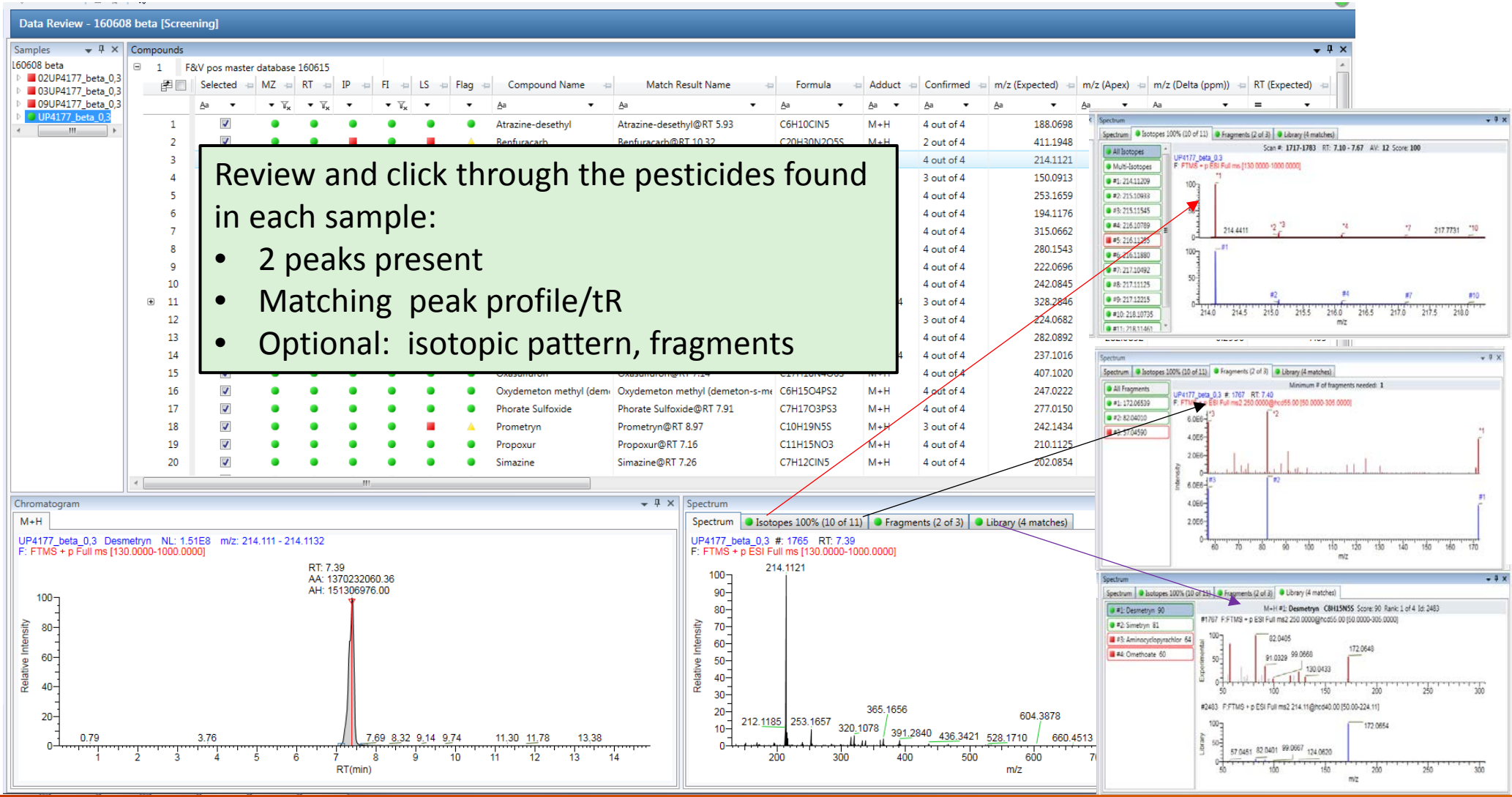
Screening Data Review for Target analyses



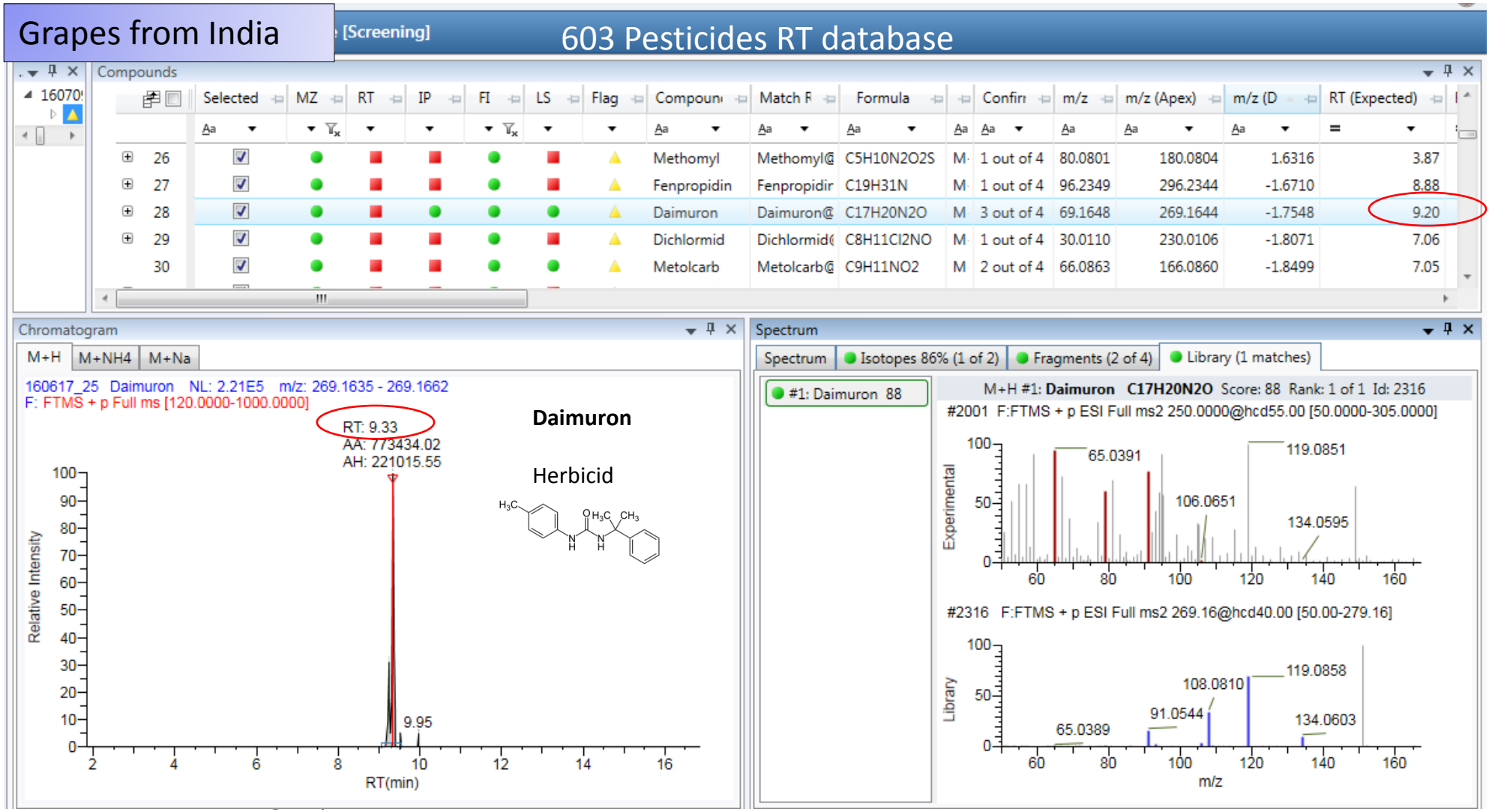
Now – let´s start with the fun part!

Finding new analytes with

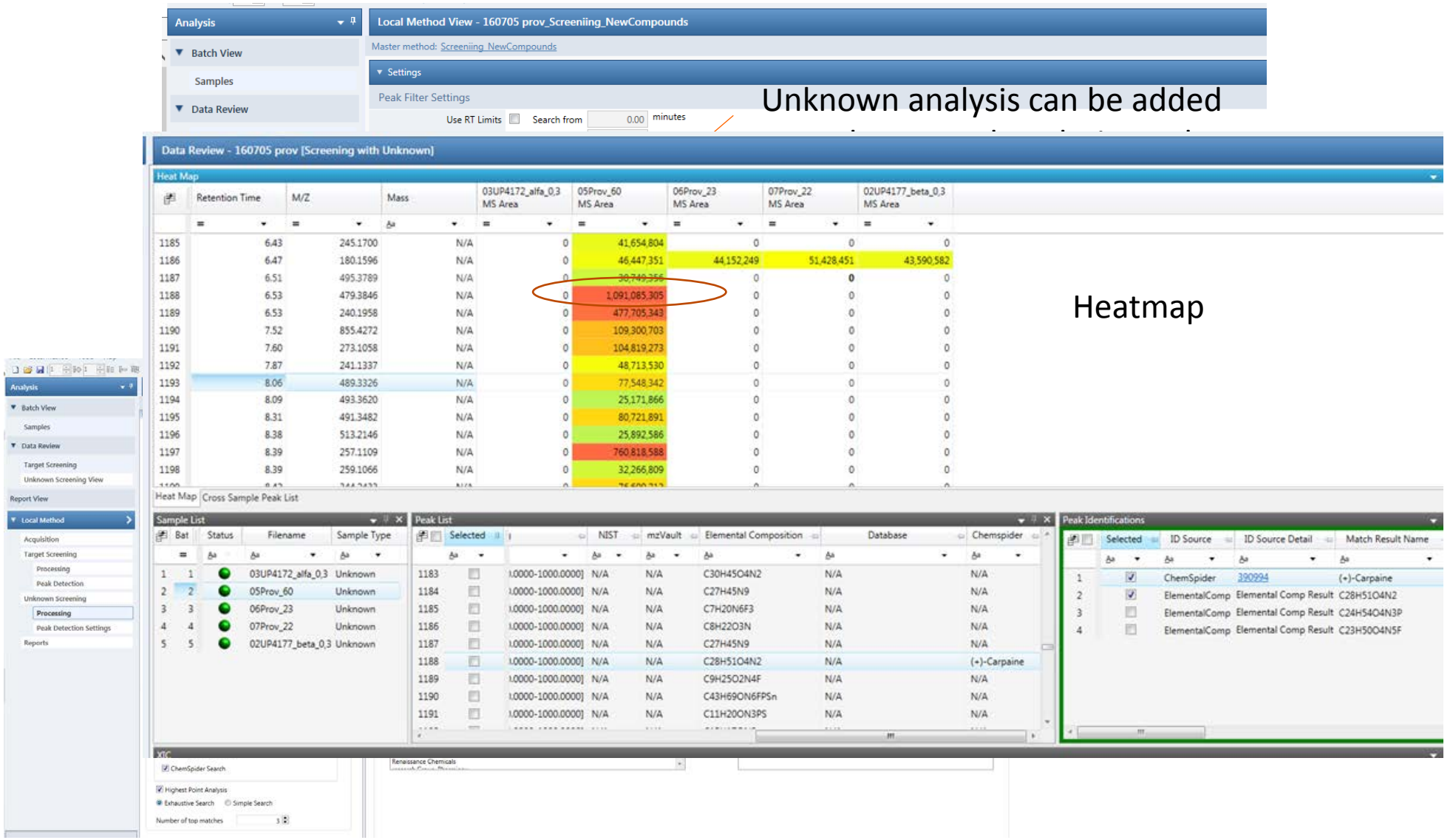
- Thermo databases
- Tracefinder unknowns

Approaches to find new pesticides; Databases & Unknown Screening

Grapes from India

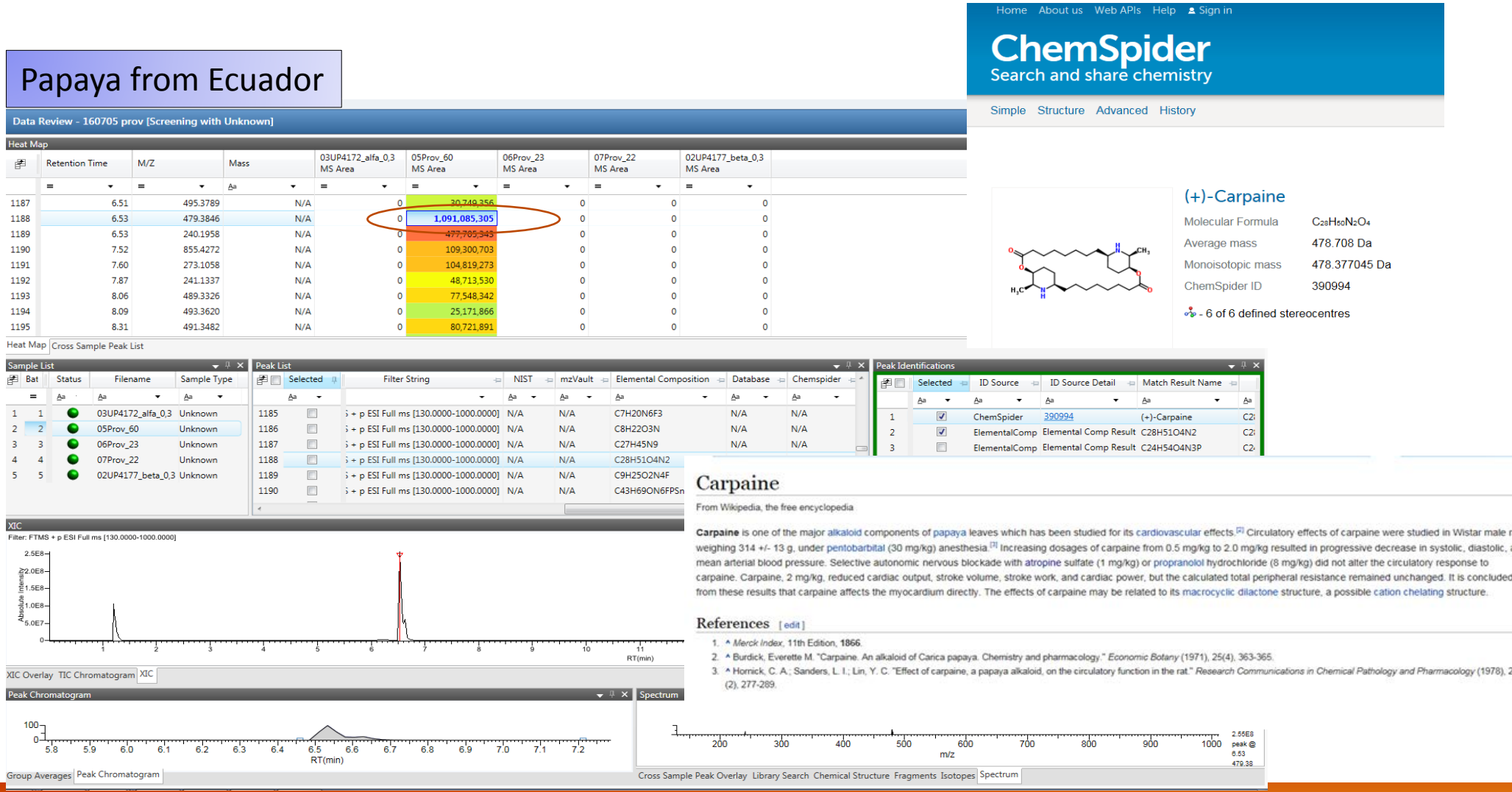


Tracefinder Unknown Screening



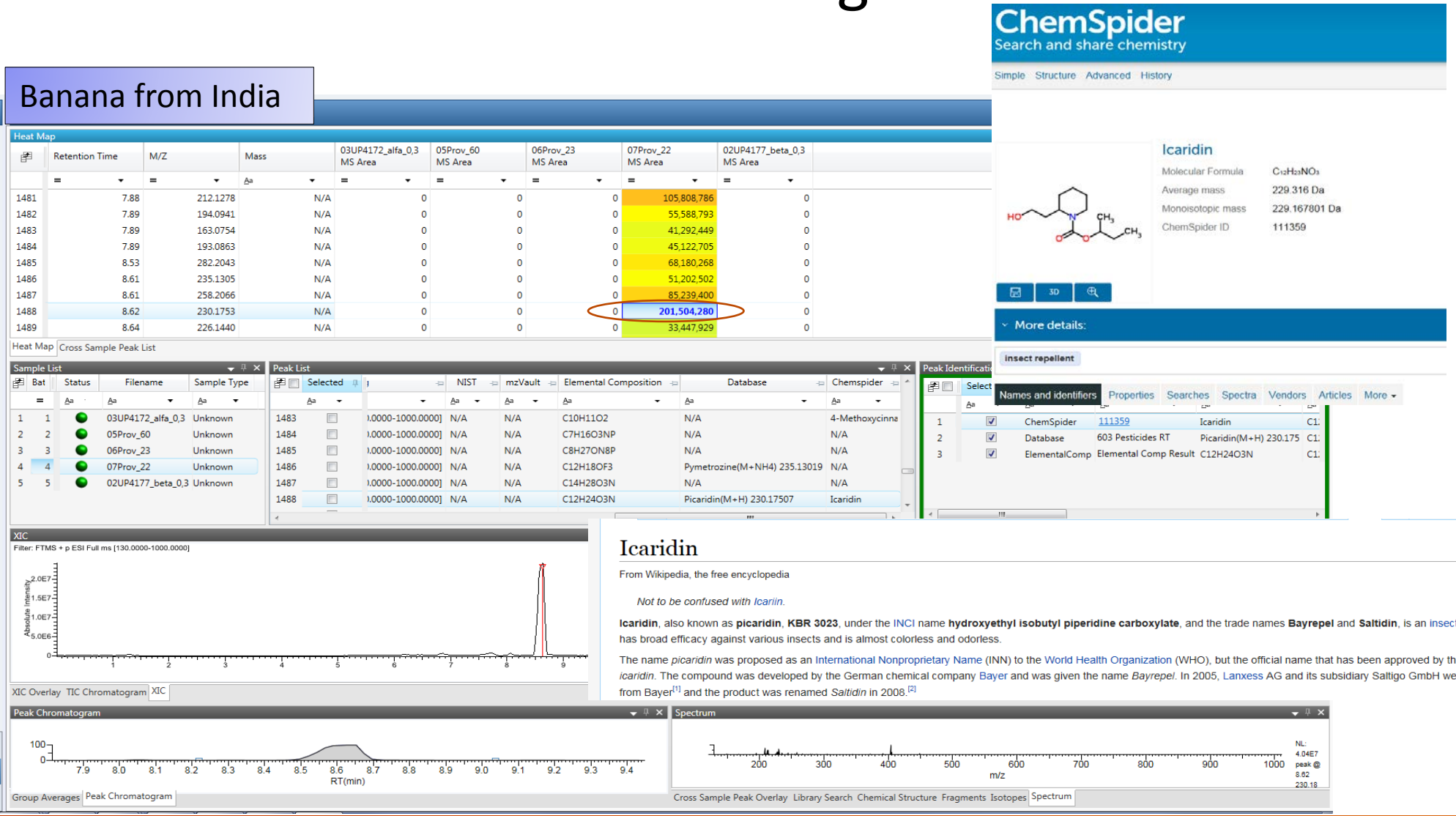
Example of a finding in papaya with Tracefinder Unknown Screening

Papaya from Ecuador

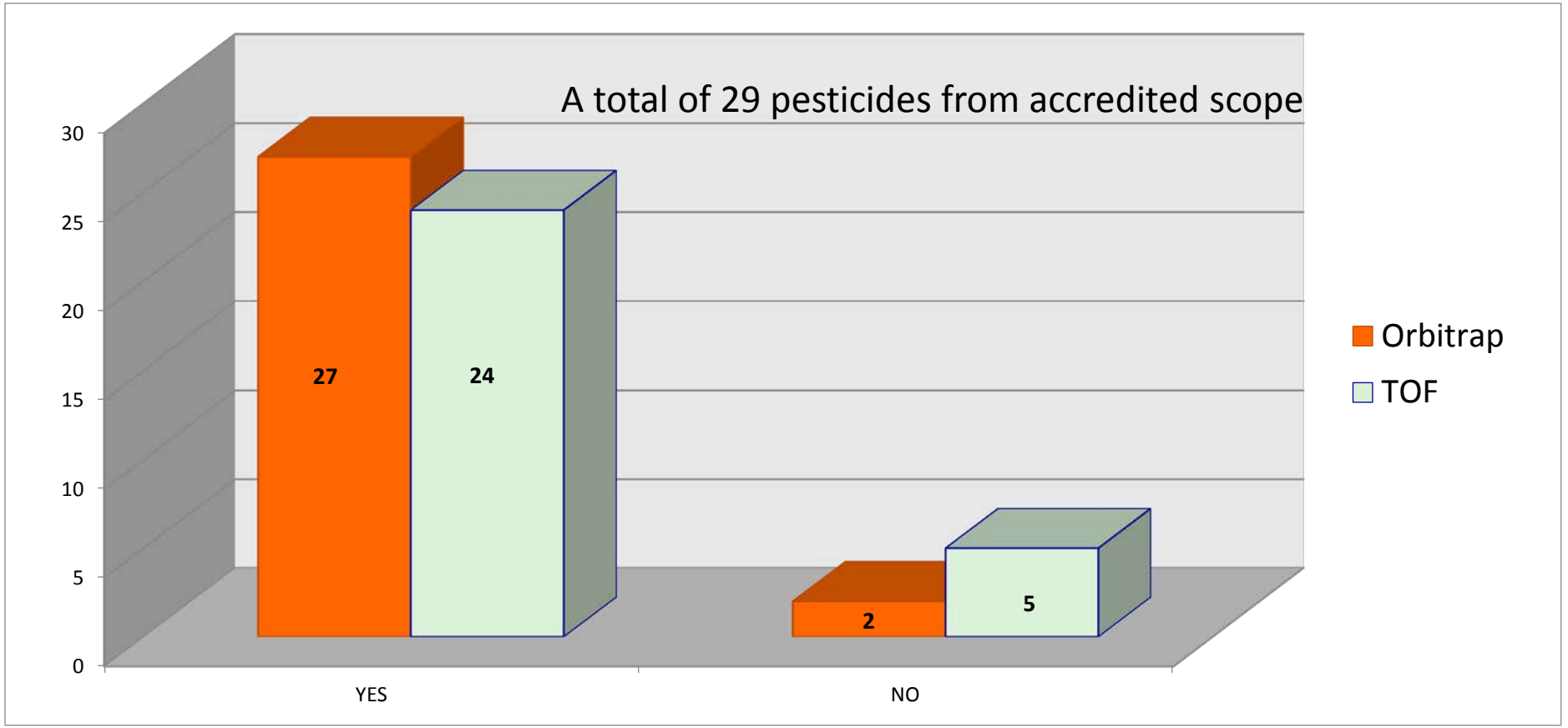


New pesticides with Tracefinder Unknown Screening

Banana from India



Orbitrap vs QTOF qualitative findings in raw water



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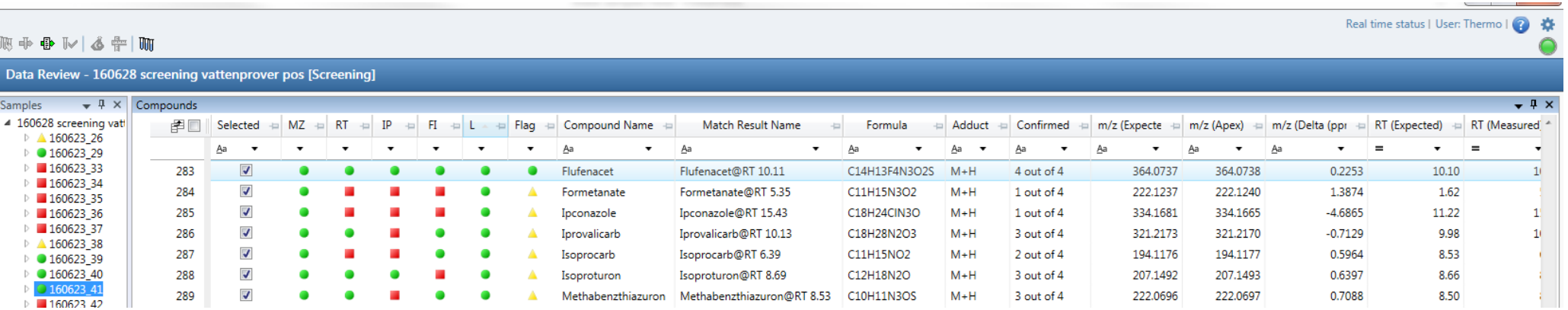
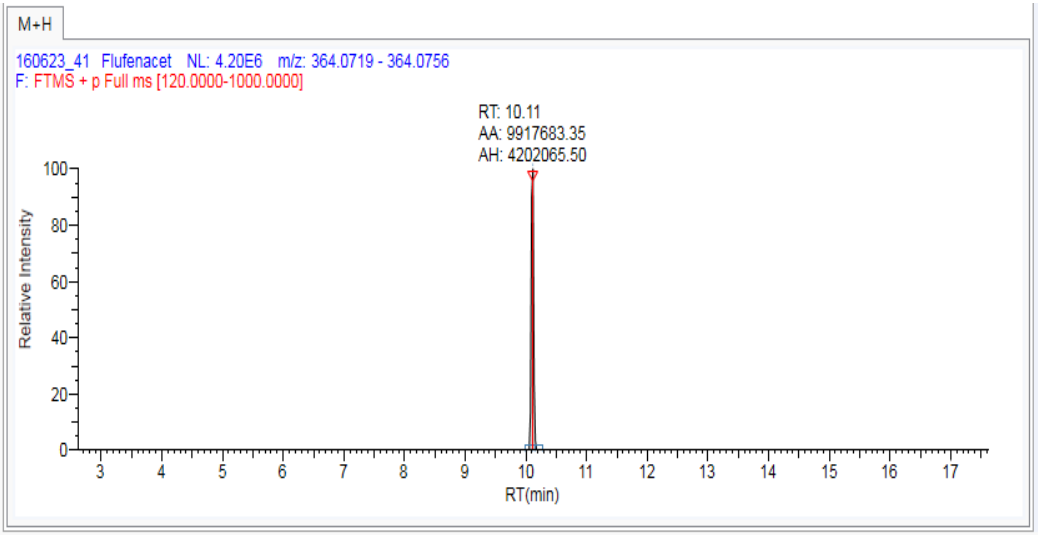
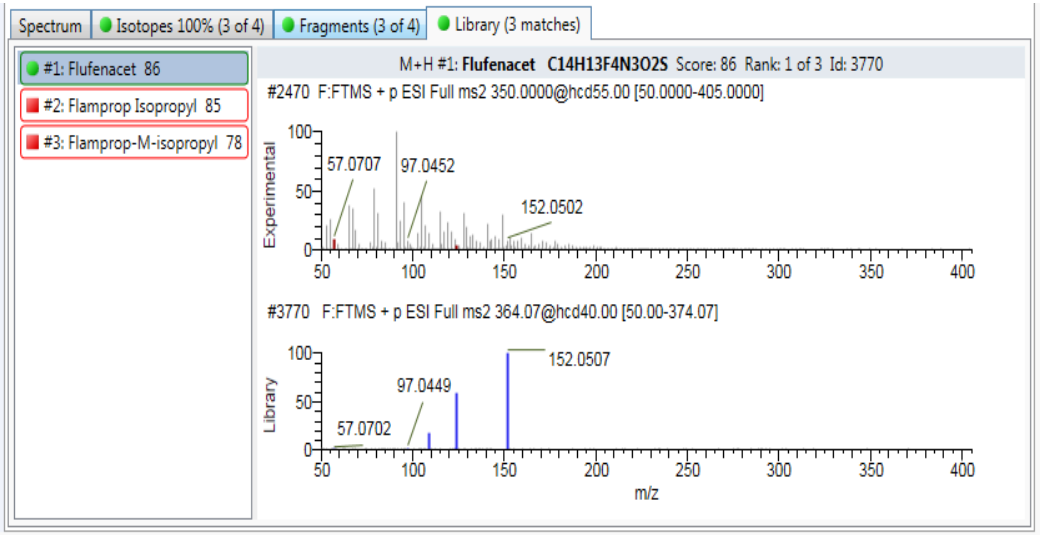
Flufenacet found by Orbitrap but not QTOF

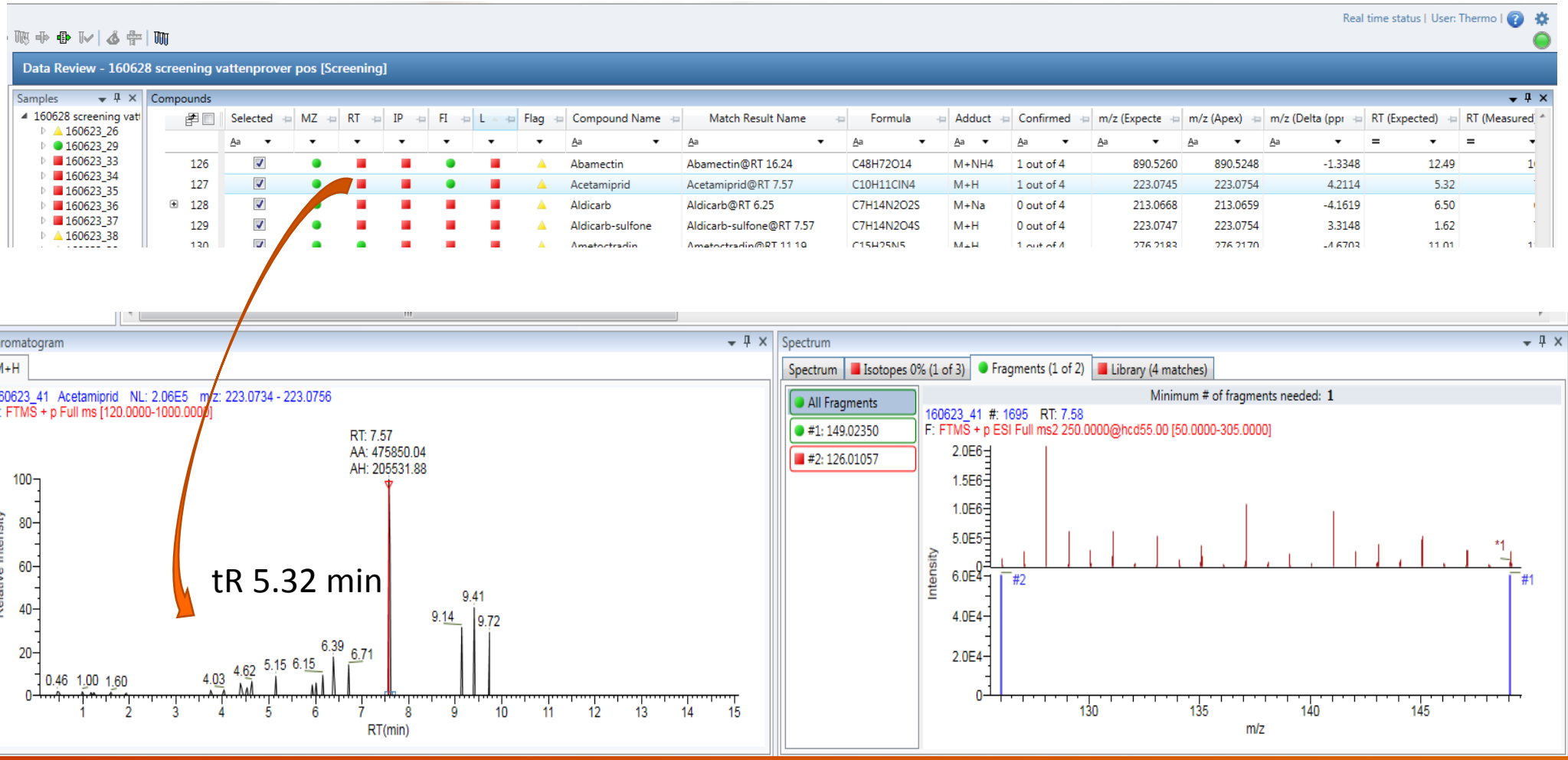


	Selected	MZ	RT	IP	FI	L	Flag	Compound Name	Match Result Name	Formula	Adduct	Confirmed	m/z (Expecte	m/z (Apex)	m/z (Delta (ppr	RT (Expected)	RT (Measured
283	☑	●	●	●	●	●	●	Flufenacet	Flufenacet@RT 10.11	C14H13F4N3O2S	M+H	4 out of 4	364.0737	364.0738	0.2253	10.10	1(
284	☑	●	■	■	■	●	▲	Formetanate	Formetanate@RT 5.35	C11H15N3O2	M+H	1 out of 4	222.1237	222.1240	1.3874	1.62	
285	☑	●	■	■	■	●	▲	Ipconazole	Ipconazole@RT 15.43	C18H24ClN3O	M+H	1 out of 4	334.1681	334.1665	-4.6865	11.22	1
286	☑	●	●	■	●	●	▲	Iprovalicarb	Iprovalicarb@RT 10.13	C18H28N2O3	M+H	3 out of 4	321.2173	321.2170	-0.7129	9.98	1(
287	☑	●	■	■	●	●	▲	Isoprocarb	Isoprocarb@RT 6.39	C11H15NO2	M+H	2 out of 4	194.1176	194.1177	0.5964	8.53	
288	☑	●	●	●	■	●	▲	Isoproturon	Isoproturon@RT 8.69	C12H18N2O	M+H	3 out of 4	207.1492	207.1493	0.6397	8.66	
289	☑	●	●	■	●	●	▲	Methabenzthiazuron	Methabenzthiazuron@RT 8.53	C10H11N3OS	M+H	3 out of 4	222.0696	222.0697	0.7088	8.50	

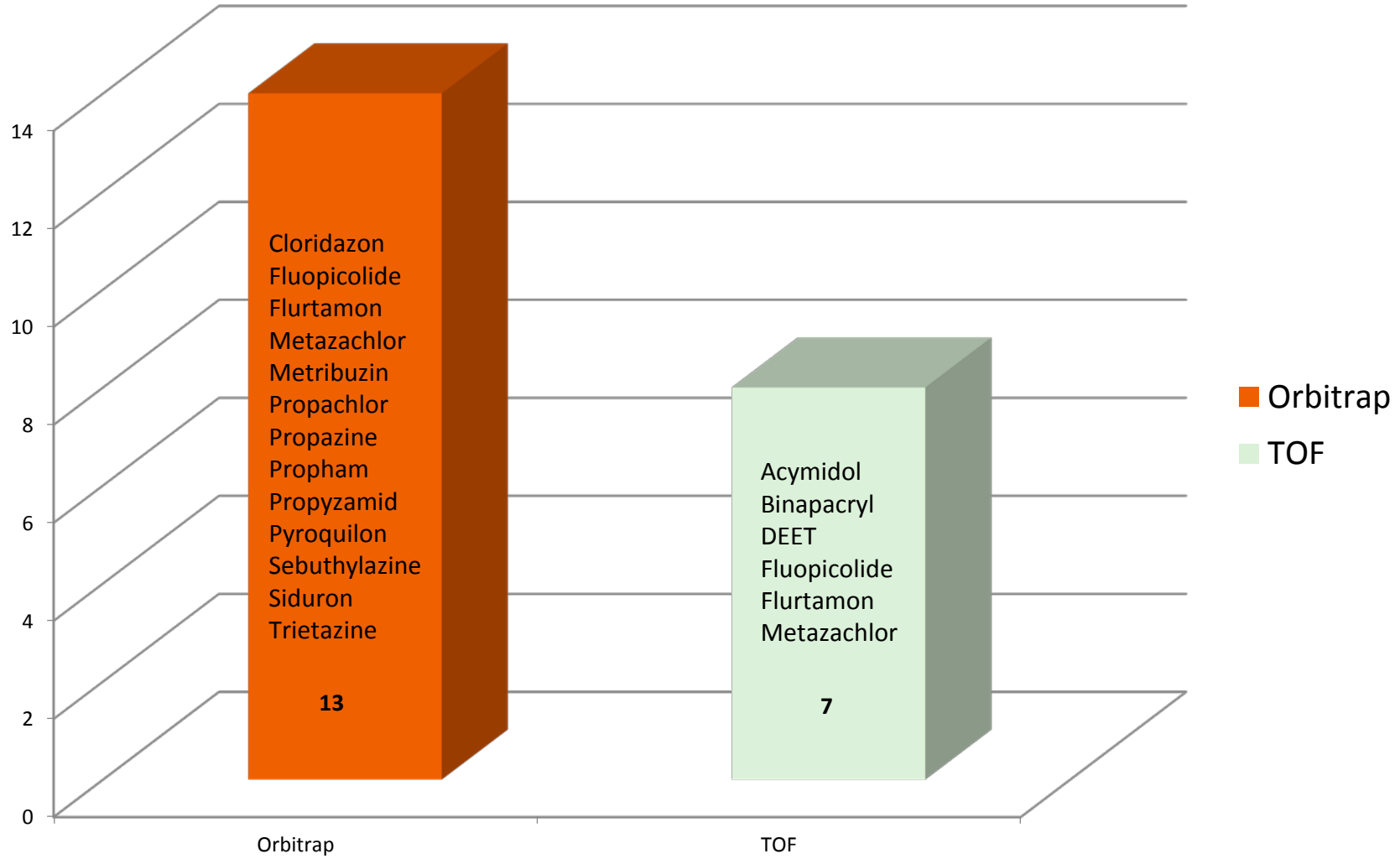




Acetamiprid found by QTOF but not Orbitrap



Tentative hits



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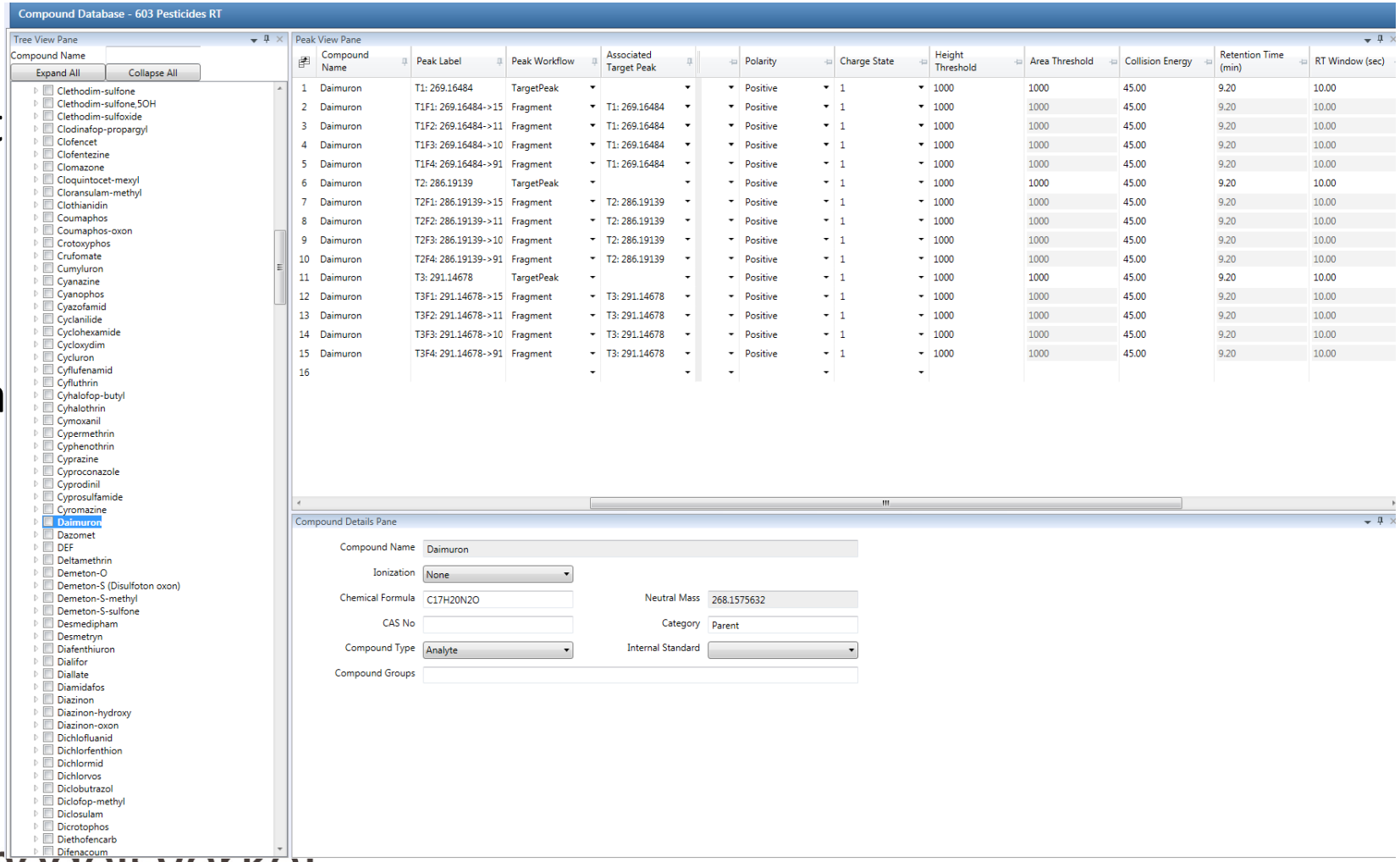
Databases for screening with tR

- Pest
- 603

Databa



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Future plans and developments

Targeted - Tracefinder

- Monitoring of pesticides including search for unknown
- Mycotoxins in cereals
- Toxins in drinking water
- Veterinary drugs in animal origin
- Alkaloids, acryl amide

Untargeted – Compound Discoverer

- Food Fraud
- Emerging risks

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Conclusions, Pros and Cons

- \+ Two techniques on one platform
- \+ Retrospective analyses – unconditional possibilities
- \+ Userfriendly, workflow as triplequads
- \+ Shared databases
- \+ Affordable

- — More work on databases with correct tR

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Acknowledgment

Pesticide-group at NFA and Heidi Pekar

World wide Orbitrap family

- Hans Mol & Paul Zomer (Rikilt, Holland)
- Łukasz Rajski (Univ of Almeria, Spain)
- Jana Hajslova & Zybnek Dzuman (Univ of Chem and Techn, Czech rep)
- Jian Wang (Food Inspec, Canada)
- Walter Hammack (Dep of Agric, Florida, US)

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- Richard Fussel, UK, Global Marketing Manager for Food & Beverage market in Chrom and MS-group
- Frans Schoutsen, NL, Sales Support Expert LSMS and Environm and Food Safety

Thank you for your attention!

Have a nice summer!