iFunnel and triggered MRM

New tools for unmatched sensitivity and unequivocal confirmation of pesticides in food

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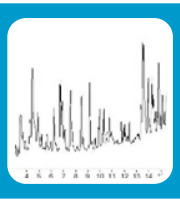
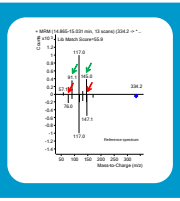
Agenda

Introduction



Validation of QQQ screening

Triggered MRM



Quan-Qual Workflow

Real world examples

Pesticide residues in food and feed

Introduction

- The protection of crops from pests and diseases requires the use of pesticides
- Pesticide residues in food and feed are possible and not necessarily illegal
- Tolerable concentrations are regulated e.g. in European Regulation (EG) No. 396/2005 or in the American Federal Regulations 40 CFR Part 180
- For food and feed produced or marketed in Europe there are maximum residue limits (MRLs) for more than 170 000 matrix-pesticide combinations
- Challenge for the analysis is the large variety of different matrices and physico-chemical properties of the pesticides
- Typical analytical methods include generic extraction (QuEChERS) and Multi-residue methods based on gaschromatography and liquid chromatography coupled to mass spectrometry which are fit for purpose
- In Europe methods are validated according to guideline SANCO/12495/2011

“Check your scope” ranking for pesticides

Published by EURL for pesticides

- Includes more than 1100 compounds
  - Pesticides currently used or used in the past
  - Pesticide metabolites of importance
- Ranking of pesticides based on:
  - Toxicological data (toxicological endpoints and endocrine disruptive activity)
  - Residue situation in crops (reporting from labs in Europe and RASFF notifications)
  - Agricultural usage (including potential for misuse and persistent pesticides)
- Current situation
  - Around 600 compounds included in routine monitoring programs
  - Only about 150 pesticides often found in food commodities

6400 Triple Quadrupole Product Line

Excellent Value and Performance

6420 Triple Quadrupole LC/MS

- Robust, Easy-to-Use
- Lowest cost of ownership
- Automate compound optimization

6430 Triple Quadrupole LC/MS

- Fast, robust,
- Targeted Protein Quantitation
- Fast polarity switching

6460 Triple Quadrupole LC/MS

- Agilent Jet Stream – sub fg sensitivity
- Fast polarity switching
- Largest mass range for a premium Triple Quad

6490 Triple Quadrupole LC/MS

- iFunnel – zeptomole sensitivity
- Curved hexapole collision cell – reduced noise
- Perfect for the most demanding applications
- Fast polarity switching

Screening and quantitation for pesticides

Use the Agilent G6490 for this application …

… to increase the number of analytes in the method to be analyzed in a single injection (even those with lower abundancies)

… to lower LOQs for pesticides in complex matrices

… to extend your analytical scope to the analysis of baby food

… to minimize matrix effects by dilution of your samples

The unique package that enables this capability is:

1. 1290 UHPLC
2. Dynamic MRM
3. Agilent Jet Stream and Dual Ion Funnel Ion sampling
4. Curved Collision cell with ion focussing and Linear acceleration for speed and sensitivity

Agilent G6490A QQQ system

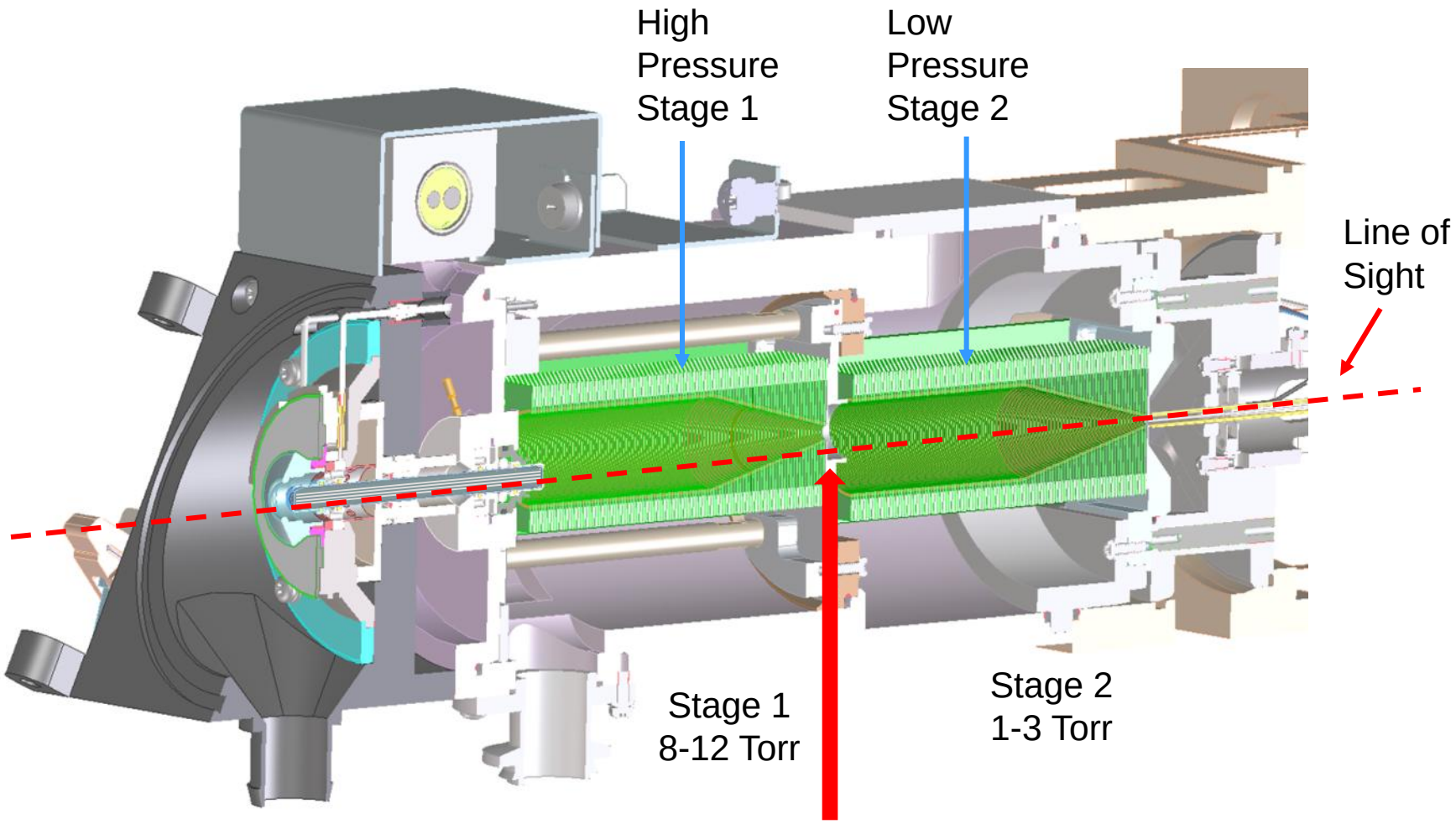
Developments for utmost sensitivity

- Ionization and Ion Transfer Technology
  - Agilent Jet Stream Ion Generation
  - Hexabore capillary
  - Dual ion funnel (iFunnel) technology
    - Two stages for ion focusing and gas removal
    - Improvements for wide *m/z* range transmission
    - Low capacitance

- Collision Cell
  - Hexapole field axial focusing curved collision cell
    - Tapered cell structure for increased ion acceptance at entrance
    - Reduced noise
- Improved Quad Drive Electronics
  - Improved Quad DC frequency response
  - Higher RF power capability
  - Quad drive frequency increased to 1.4 MHz

Agilent iFunnel technology

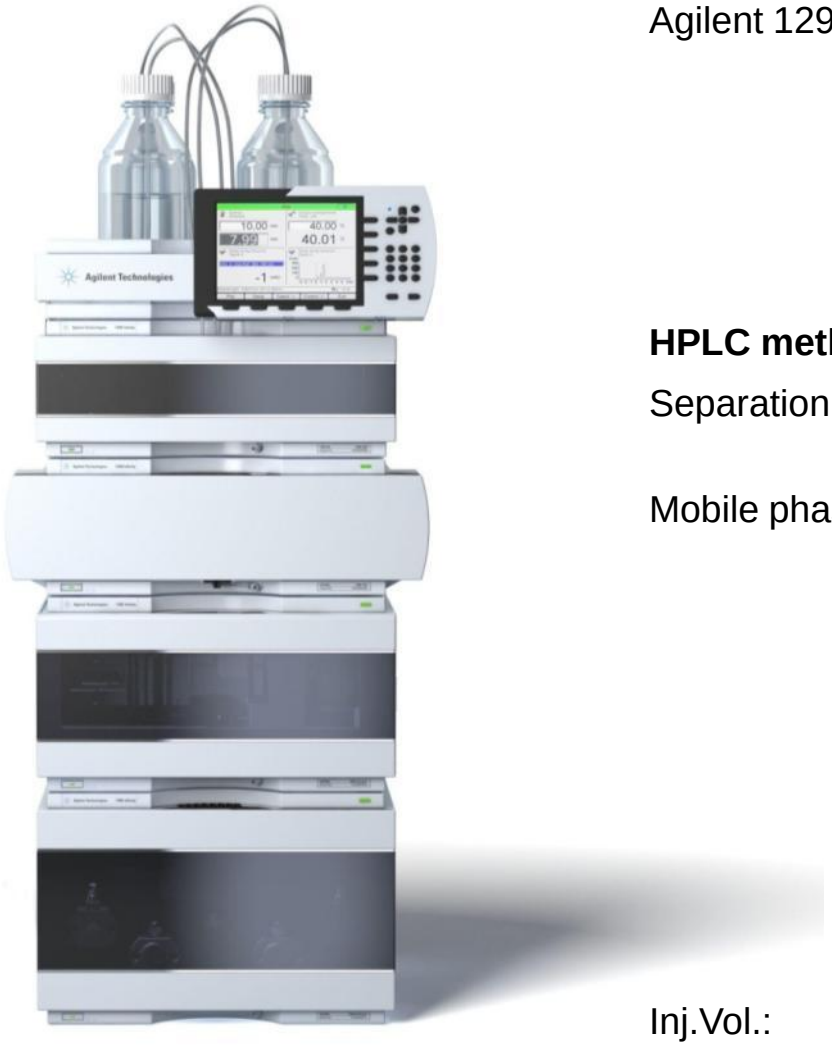
Two stage ion funnel manages the gas load



Offset ion funnels to prevent neutrals from going straight through to MS

Multi residue pesticide method

HPLC method



Agilent 1290 Infinity LC system consisting of:

- binary pump
- wellplate sampler
- column compartment
- diode array detector (not used)

HPLC method

Separation column:	ZORBAX Eclipse Plus C-18 RRHD column, 100 x 2.1 mm, 1.8 µm @ 25°C	
Mobile phase:	A: 5 mM ammonium formate	
	B: methanol + 5 mM ammonium formate	
	Flow:	0.6 ml/min
	Gradient:	0.00 min 5 % B
		0.20 min 5 % B
		2.20 min 50 % B
		10.50 min 100 % B
		13.00 min 100 % B
		13.10 min 5 % B
		15.00 min 5 % B
Inj.Vol.:	2 µl	

Multi residue pesticide method

MS method

Spray chamber conditions:

Gas temp.:	120°C
Dry gas:	15 l/min
Nebulizer:	35 psi
Sheath gas temp:	375°C
Sheath gas flow:	12 l/min

	Positive	**Negative**
CapVoltage:	3500 V	3000 V
Nozzle voltage	300 V	500 V

Automatic setup of MRM tables based on selected cycle time, retention times and retention time windows for the individual compounds

- Cycle time 600 ms
- Interscan delay 3.5 ms
- Total No. of MRMs 635
- Maximum No. Of concurrent MRMs 74
- Minimum Dwell time 4.61 ms
- Maximum Dwell time 296.5 ms

Multi residue pesticide method

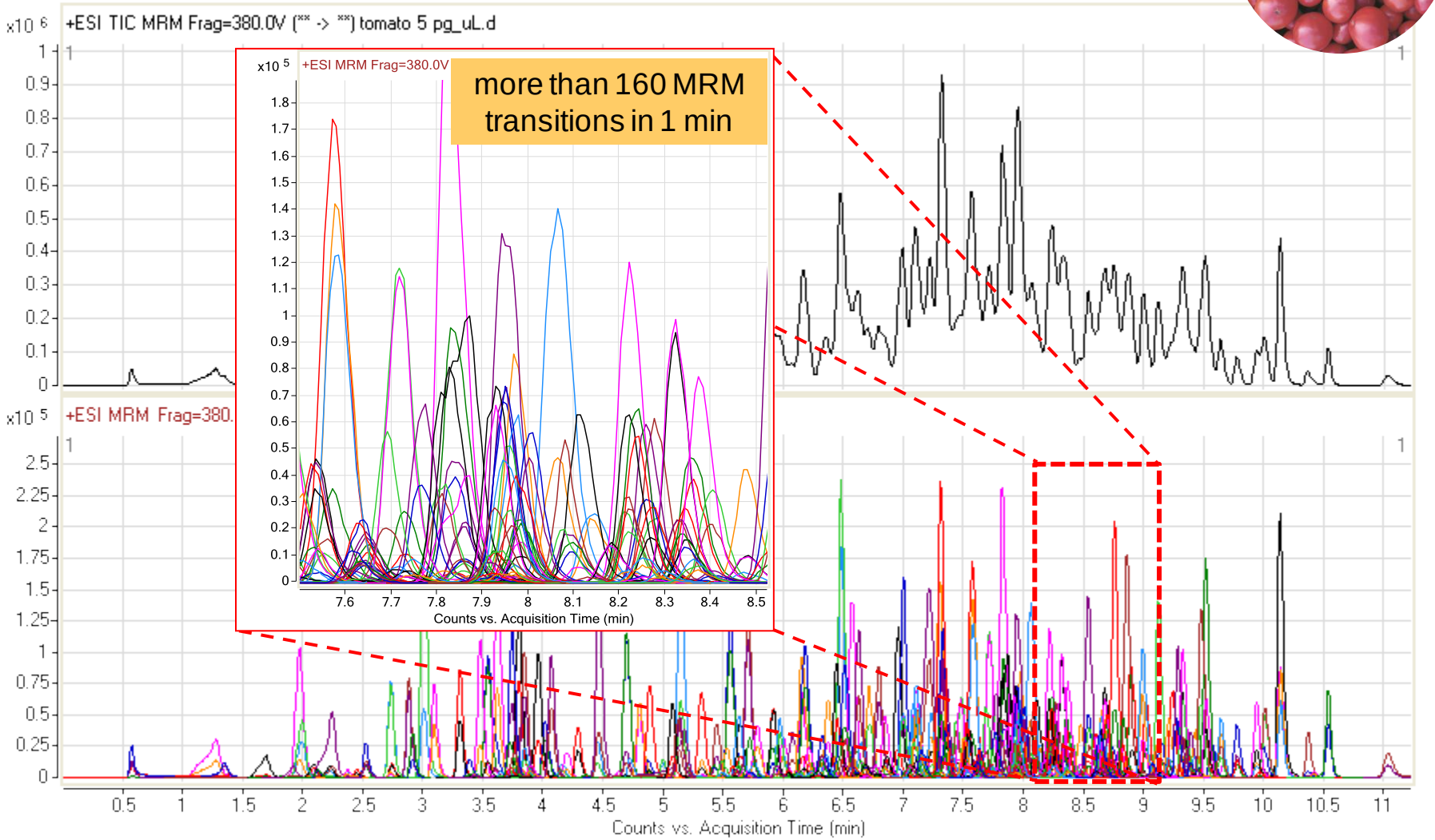
Compounds included in method

Acephat	Butocarboxim-sulfoxid	Desmedipham	Fenamiphos-sulfon	Flusilazol	Metamitron	Oxadiazon	Proquinazid	Terbutylazin
Acetamiprid	Buturon	DMSA	Fenamiphos-sulfoxid	Flutolanil	Metazachlor	Oxadixyl	Prosulfocarb	Tetraconazol
Acrinathrin	Cadusaphos	Dichlofluanid	Fenarimol	Flutriafol	Metconazol	Oxamyl	Pymetrozin	Thiabendazol
Aldicarb	Carbaryl	Dichlorvos	Fenazaquin	Foramsulfuron	Methabenzthiazuron	Oxasulfuron	Pyraclostrobin	Thiacloprid
Aldicarb-sulfon	Carbendazim	Diclofop-methyl	Fenbuconazol	Formetanat-Hydrochlorid	Methacrifos	Oxydemeton-sulfon	Pyrazophos	Thiamethoxam
Aldicarb-sulfoxid	Carbofuran 3 hydroxy	Dicrotophos	Fenbutatinoxid	Formothion	Methamidophos	Paclobutrazol	Pyridaben	Thifensulfuron-methyl
Alloxydim	Carbofuran	Diethofencarb	Fenhexamid	Fosthiazat	Methidathion	Paraoxon-methyl	Pyridaphenthion	Thiodicarb
Amidosulfuron	Carbosulfan	Difenoconazol	Fenobucarb	Fuberidazol	Methiocarb	Penconazol	Pyridat	Thiofanox sulfon
Aminopyralid	Carfentrazone-ethyl	Difenoxuron	Fenoxaprop free acid	Furathiocarb	Mthiocarb-sulfon	Pencycuron	Pyrifenox	Thiofanox sulfoxid
Amitraz	Chlorantraniliprol	Diflubenzuron	Fenoxycarb	Halosulfuron-methyl	Methiocarb-sulfoxid	Pendimethalin	Pyrimethanil	Thiofanox
Asulam	Chlorfenvinphos	Diflufenican	Fenpiclonil	Haloxyfop free acid	Methomyl	Pethoxamid	Pyriproxifen	Thiophanat
Atrazin	Chlorfluazuron	Dimefuron	Fenpropimorph	Hexaconazol	Methoxyfenozid	Phenmedipham	Pyroxsulam	Thiophanat-methyl
Avermectin B1a Metabolite	Chloridazon (Pyrazon)	Dimethenamid	Fenpyroximat	Hexaflumuron	Metobromuron	Phenthoat	Quinalphos	Tolclophos-methyl
Avermectin B1a	Chlorimuron-ethyl	Dimethoat	Fenthion	Hexythiazox	Metolachlor	Phorat	Quinmerac	Tolylfluanid
Avermectin B1b	Chloroxuron	Dimethomorph	Fenthion-oxon	Imazalil	Metosulam	Phosalon	Quinoxyfen	Topramezone
Azimsulfuron	Chlorsulfuron	Dimoxystrobin	Fenthion-oxon-sulfon	Imidacloprid	Metoxuron	Phosmet	Quizalfop free acid	Tralkoxydim
Azinphos-ethyl	Chlozolinat	Diniconazol	Fenthion-oxon-sulfoxid	Indoxacarb	Metrafenon	Phosmet-oxon	Quizalofop-ethyl	Triadimefon
Azinphos-methyl	Chromafenozid	Dioxathion	Fenthion-sulfon	Ipconazol	Metribuzin	Phosphamidon	Rimsulfuron	Triadimenol
Azoxystrobin	Clethodim	Diuron	Fenthion-sulfoxid	Iprodion	Metsulfuron-methyl	Phoxim	Rotenone	Triasulfuron
Beflubutamid	Clofentezin	EPN	Flazasulfuron	Iprovalicarb	Mevinphos	Picoxystrobin	Sethoxydim	Triazofos
Benalaxyl	Clomazon	Epoxyconazol	Flonicamid	Isoproturon	Molinat	Pinoxaden	Siduron	Tribenuron-methyl
Benfuracarb	Clopyralid	Ethaboxam	Florasulam	Isoxaflutol	Monocrotophos	Piperonyl butoxid	Silthiopham	Trichlorfon
Bensulfuron-methyl	Clothianidin	Ethiofencarb	Fluazifop free acid	Kresoxim methyl	Monolinuron	Pirimicarb	Simeconazol	Tricyclazol
Benthiavalicarb	Cyazofamid	Ethiofencarb-sulfon	Fluazifop-P-butyl	Lenacil	Monuron	Pirimiphos-methyl	Spinosad	Trifloxystrobin
Bifenazat	Cycloxydim	Ethiofencarb-sulfoxid	Flufenacet	Linuron	Myclobutanil	Pirmicarb-desmethyl	Spirotetramat	Triflumizol
Bifenox	Cyhexatin	Ethion	Flufenoxuron	Lufenuron	Napropamid	Prochloraz	Spiroxamin	Triflumuron
Bispyribac	Cymoxanil	Ethiprol	Flumetsulam	Malaoxon	Neburon	Profenophos	Sulfosulfuron	Triflusulfuron-methyl
Bitertanol	Cyproconazol	Ethirimol	Fluometuron	Mandipropamid	Nicosulfuron	Promecarb	Tebuconazol	Triforin
Bromacil	Cyprodinil	Ethofumesat	Fluopicolid	Mecarbam	Nitenpyram	Propamocarb	Tebufenozid	Trinexapac-ethyl
Bromuconazol	Cyromazin	Ethoprophos	Fluoroglycofen-ethyl	Mepanipyrim	Novaluron	Propaquizafop	Tebufenpyrad	Triticonazol
Bupirimat	Daminozid	Etofenprox	Fluoxastrobin (E)	Mesosulfuron-methyl	Nuarimol	Propargite	Teflubenzuron	Tritosulfuron
Buprofezin	DEET	Famoxadon	Fluquinconazol	Mesotrione	Ofurace	Propiconazol	Tembotrion	Zoxamid
Butocarboxim	Demeton-S-methyl	Fenamidon	Fluroxypyr	Metaflumizone	Omethoat	Propoxur	Tepraloxydim	
Butoxycarboxim	Demeton-S-methyl-sulfon	Fenamiphos	Flurtamon	Metalaxyl	Orthosulfamuron	Propyzamid	Terbutryn	

313 compounds including 9 isomers

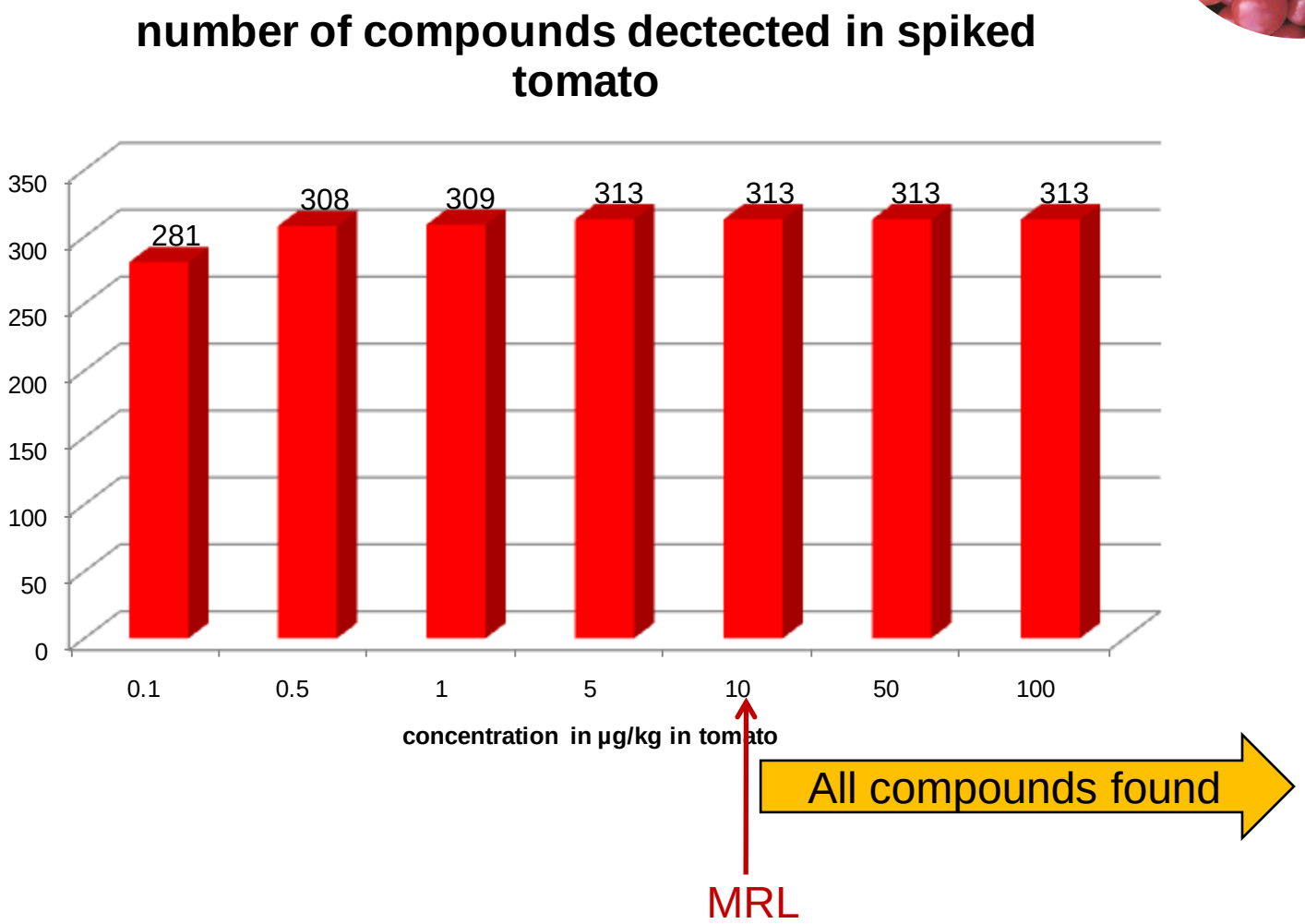
Multi residue pesticide method

MRM traces for tomato extract spiked at 10 µg/kg



Results for tomato extract

Coverage of method for tomato matrix

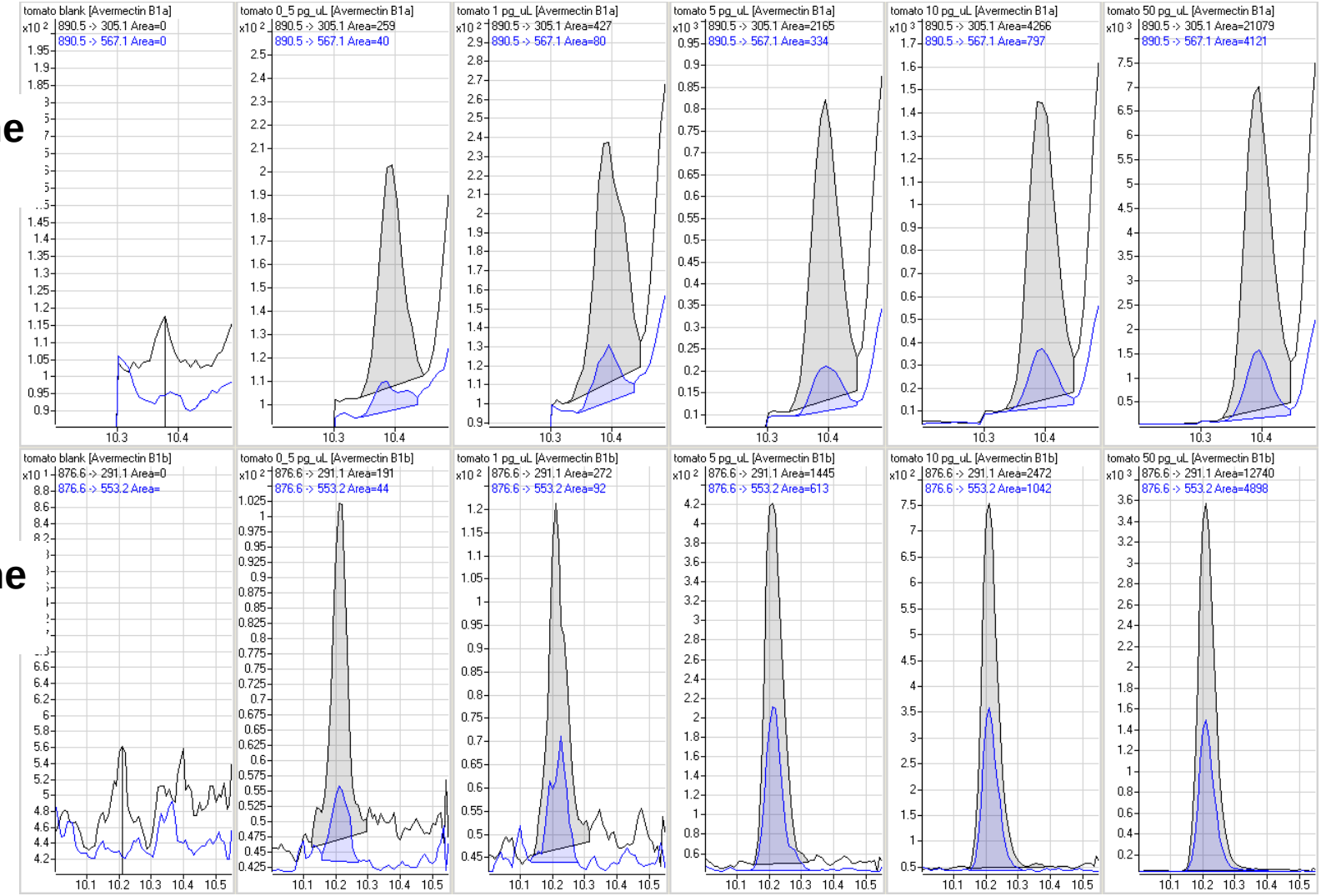


Results for tomato extract

Avermectine B1a and B1b

Avermectine B1a

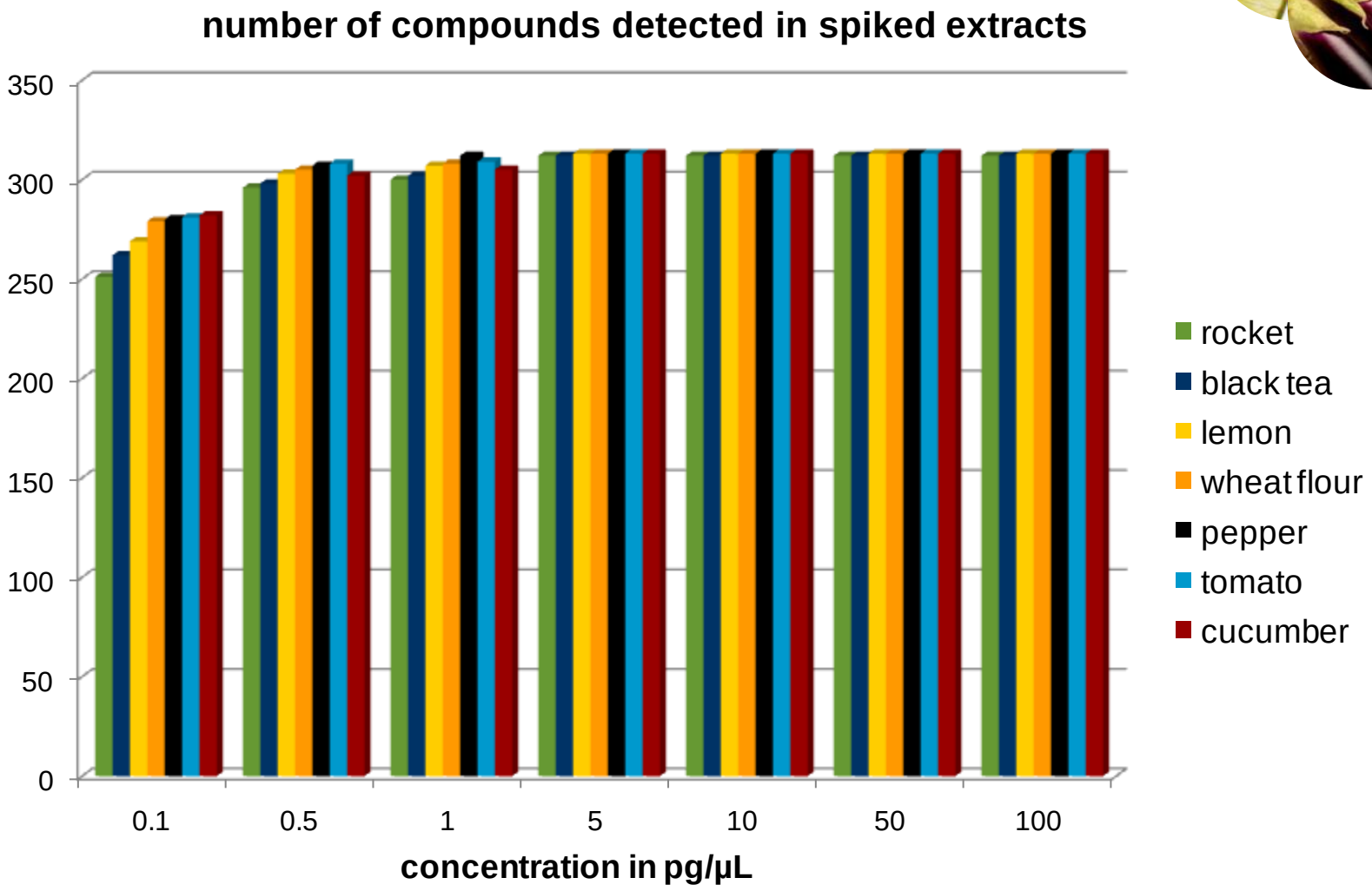
Avermectine B1b



blank | 0.5 µg/kg | 1 µg/kg | 5 µg/kg | 10 µg/kg | 50 µg/kg

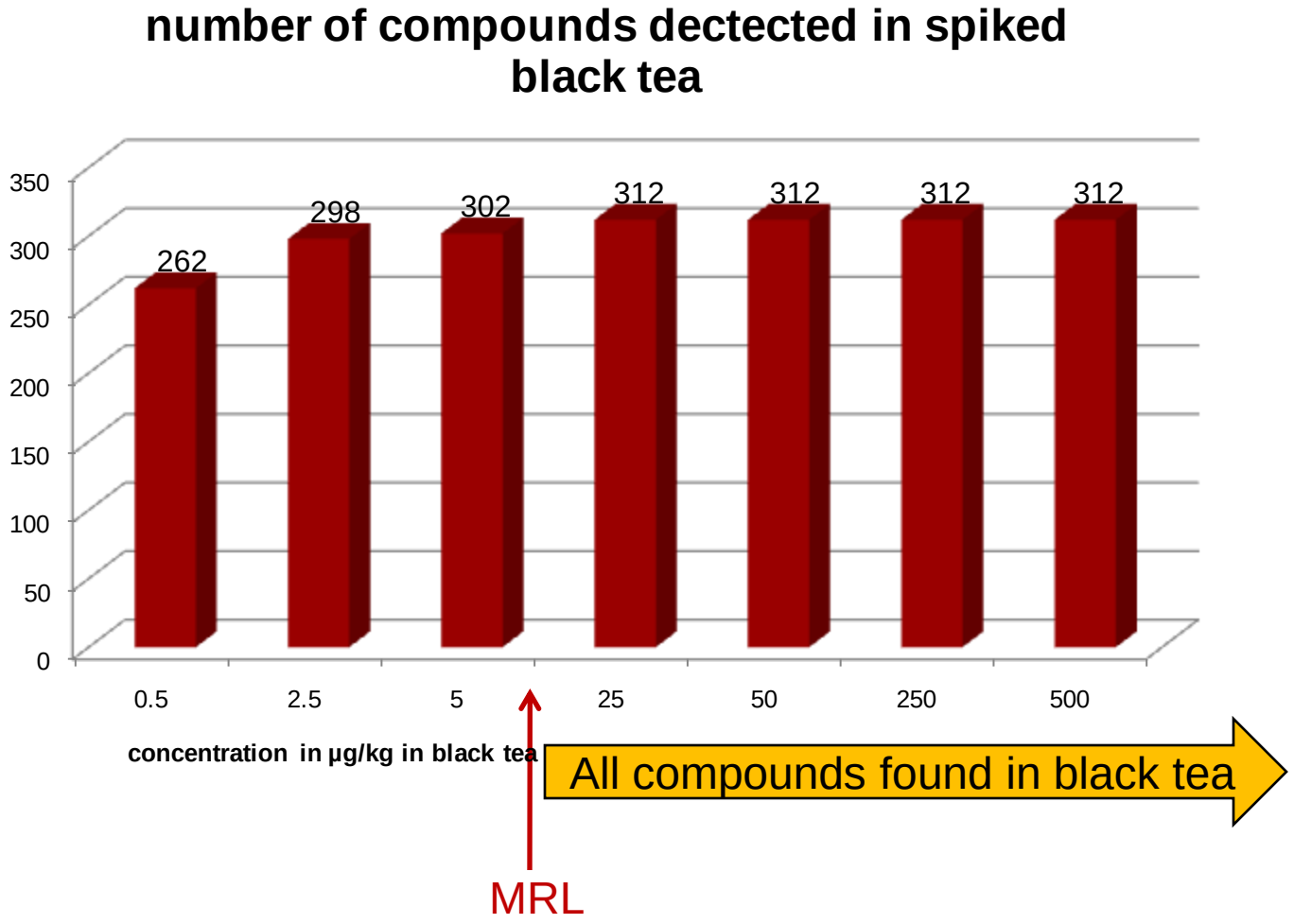
Results for all matrices

Coverage of method for different matrices



Results for black tea

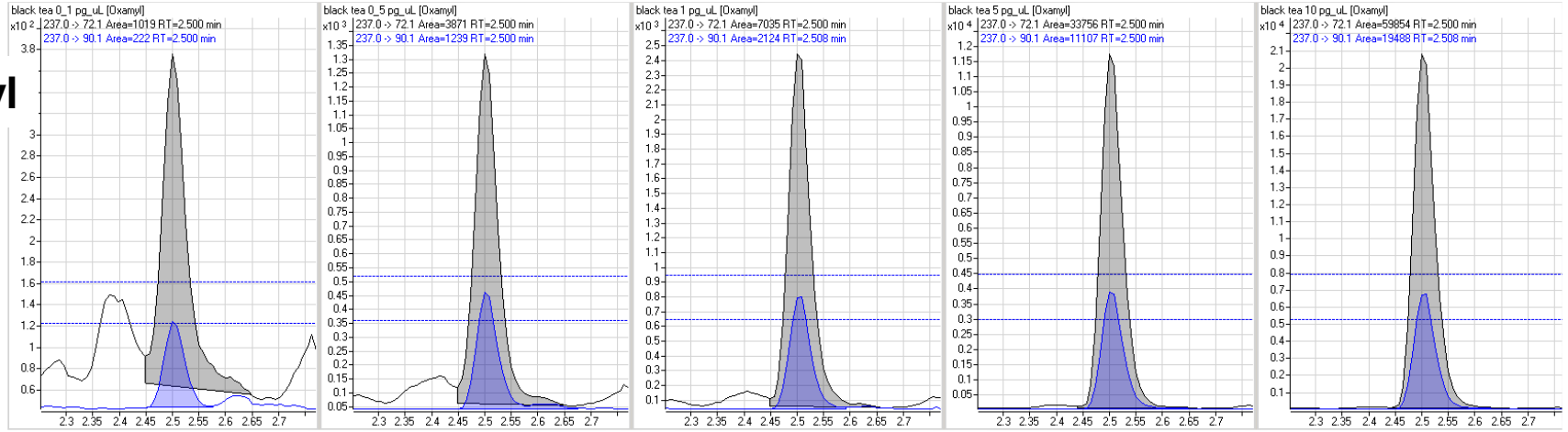
Coverage of method for black tea



Results for black tea

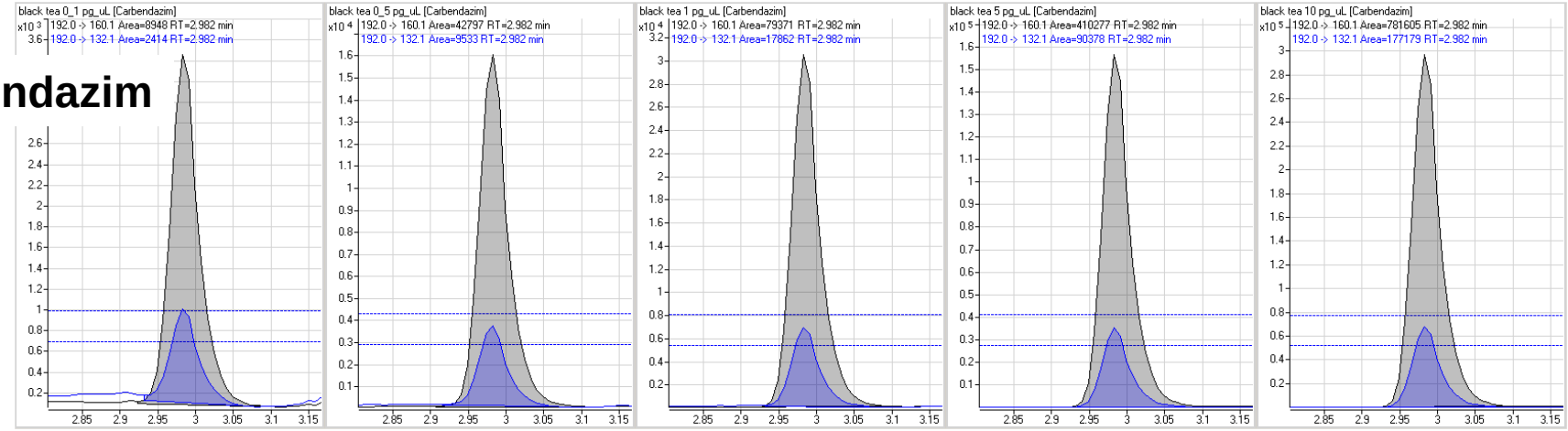
Oxamyl and Carbendazim (High in “Check Your Scope” ranking)

Oxamyl



0.5 µg/kg | 2.5 µg/kg | 5 µg/kg | 25 µg/kg | 50 µg/kg

Carbendazim

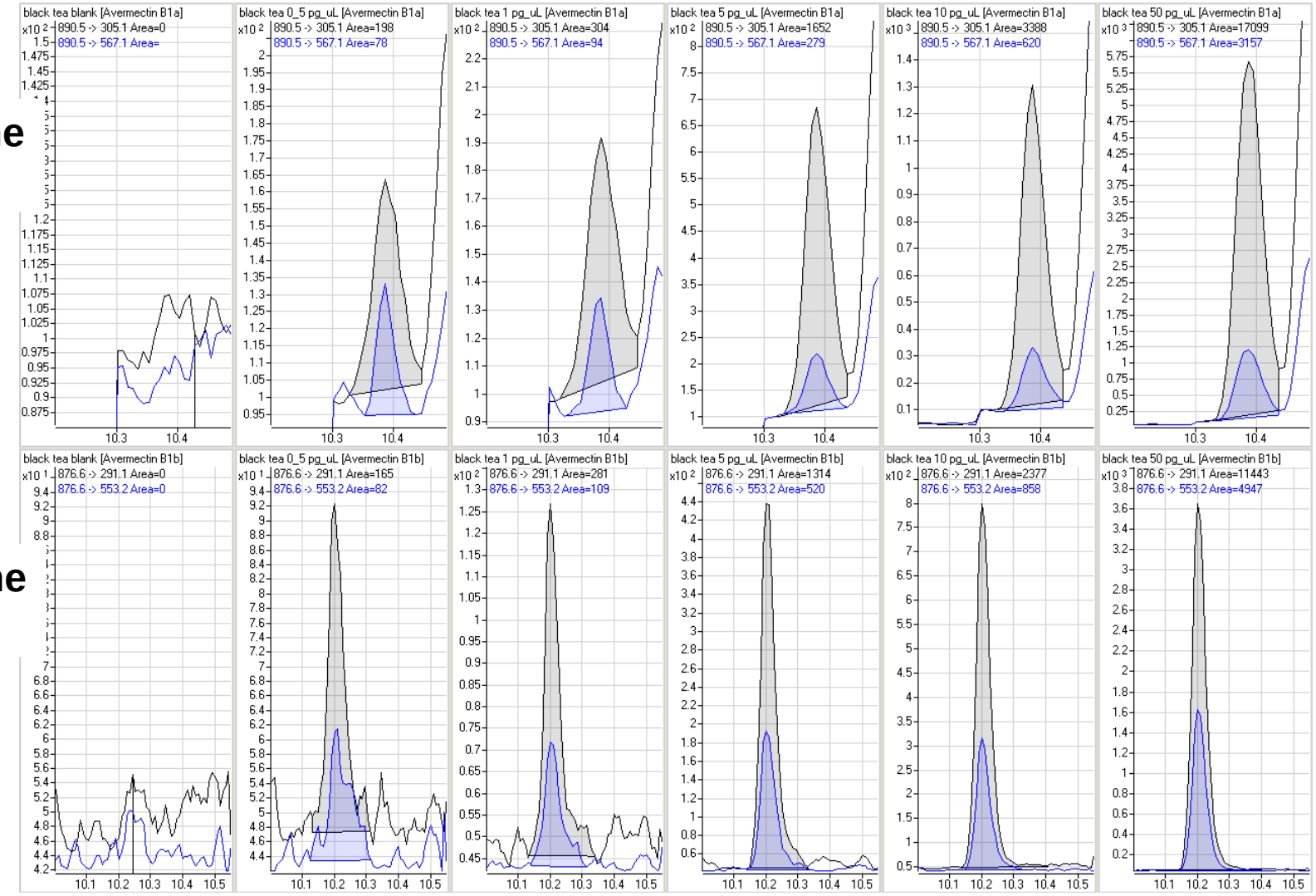


Results for black tea

Avermectine B1a and B1b

Avermectine B1a

Avermectine B1b

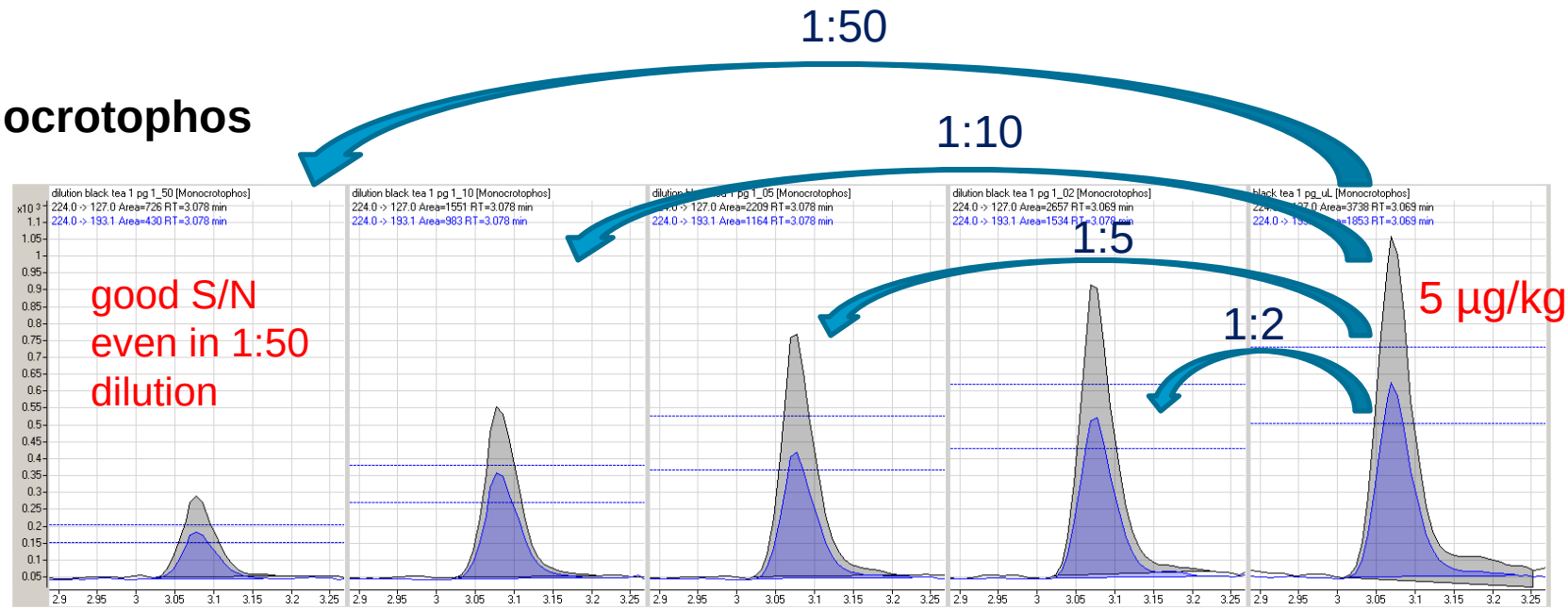


blank | 2.5 µg/kg | 5 µg/kg | 25 µg/kg | 50 µg/kg | 250 µg/kg

Dilution of black tea extract

Results for monocrotophos

Monocrotophos

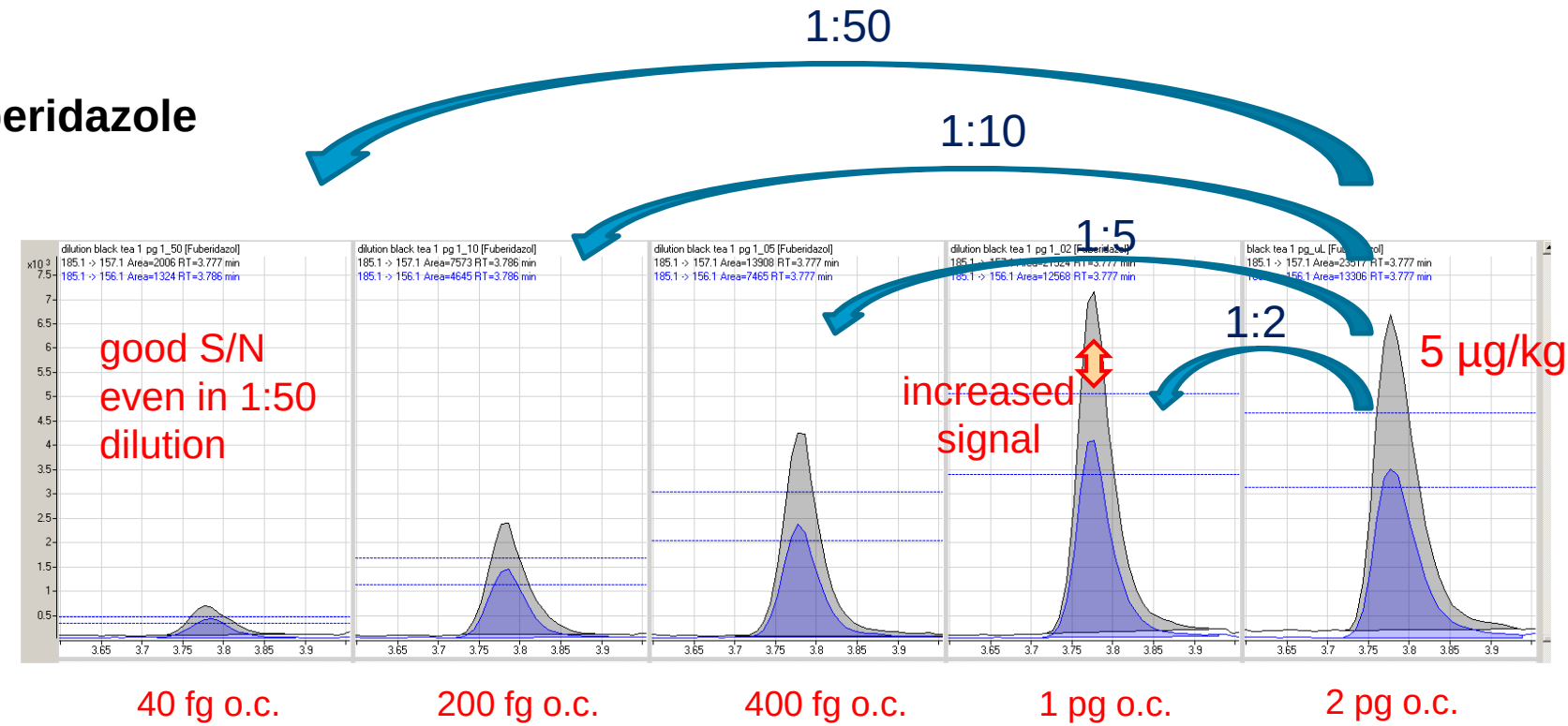


40 fg o.c. | 200 fg o.c. | 400 fg o.c. | 1 pg o.c. | 2 pg o.c.

Dilution of black tea extract

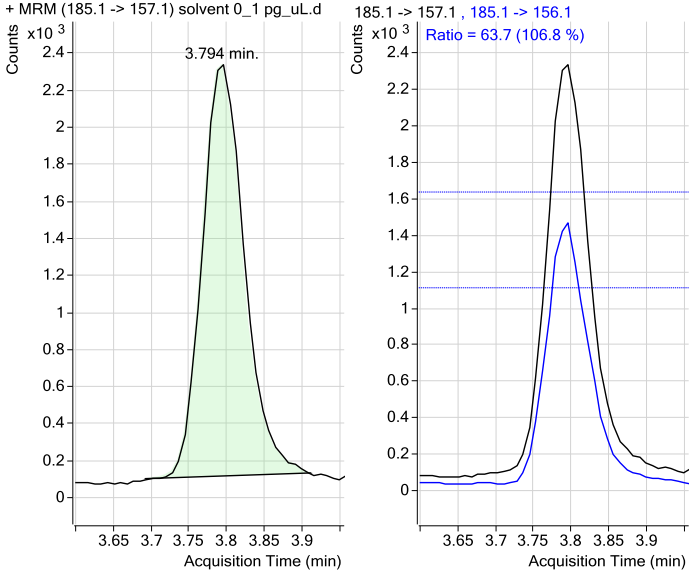
Results for fuberidazole

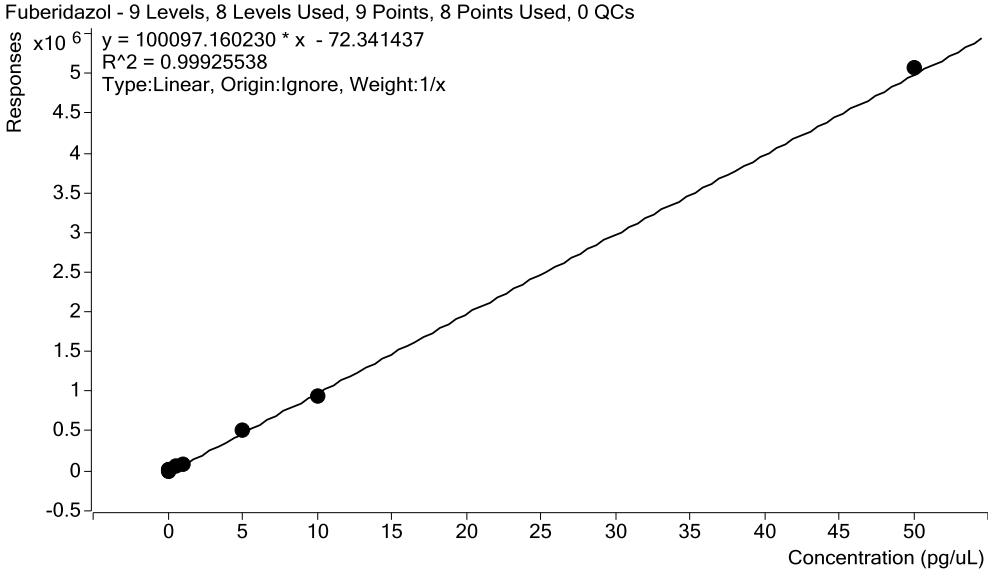
Fuberidazole



Results for Fuberidazole

Calibration in solvent and recoveries in different matrices

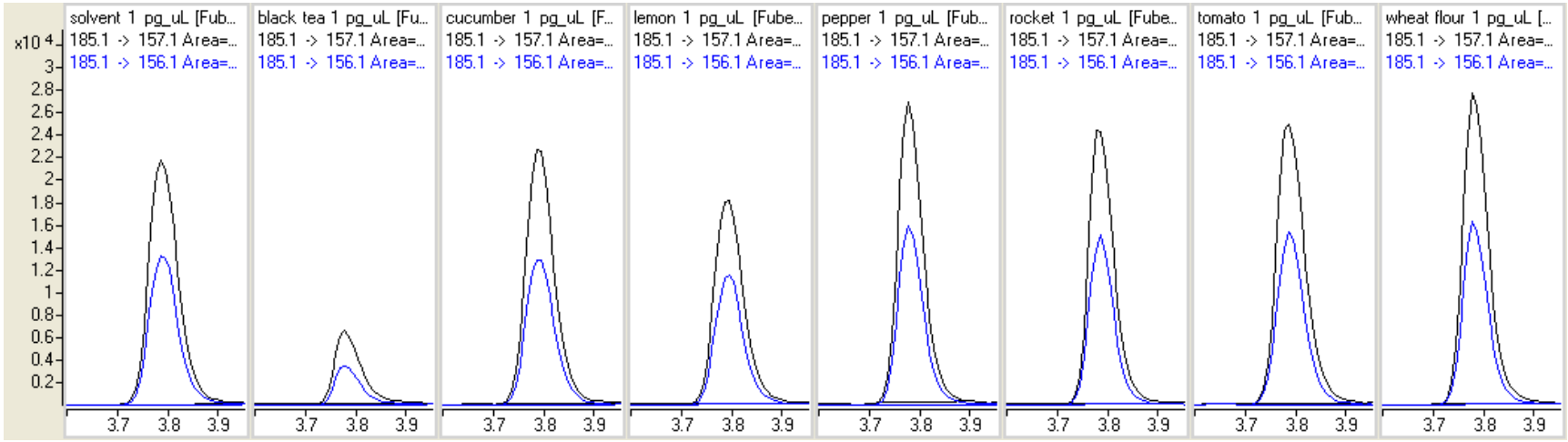




- Quantifier: 185.1 → 157.1
- Qualifier: 185.1 → 156.1
- Peak width: 16.2 s
- Cycle time: 600 ms
- Data points: 27

Results for Fuberidazole

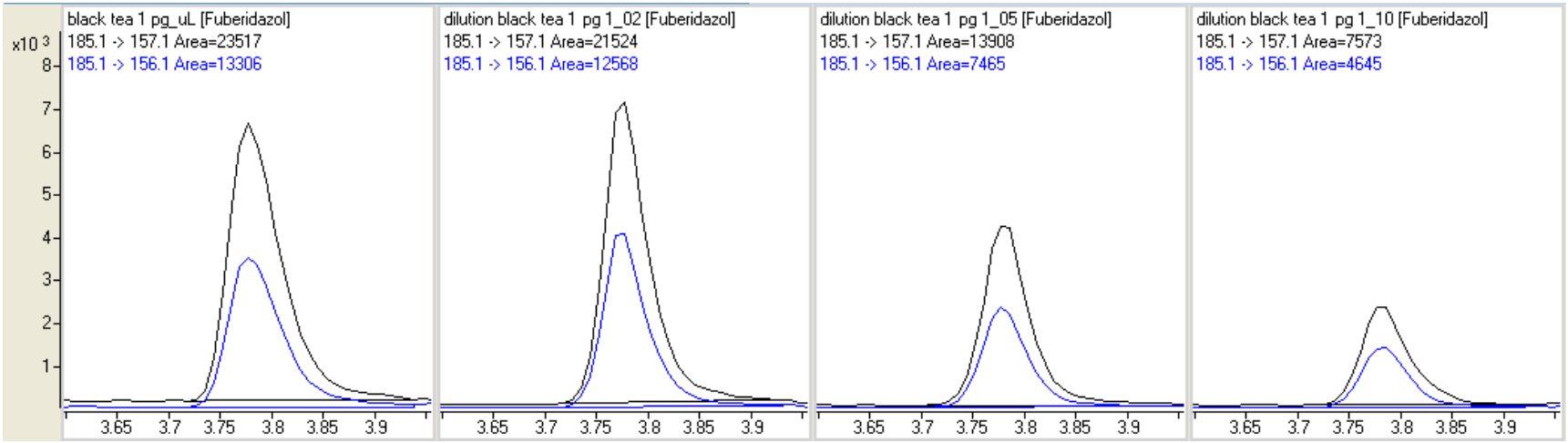
Calibration in solvent and recoveries in different matrices



Matrix	Concentration	Recovery
Solvent	0.94	
Black tea	0.25	24.6
Cucumber	0.95	94.7
Lemon	0.78	78.4
Pepper	0.97	97.0
Rocket	0.89	89.8
Tomato	1.04	104.1
Wheat flower	1.04	103.5

Results for Fuberidazole

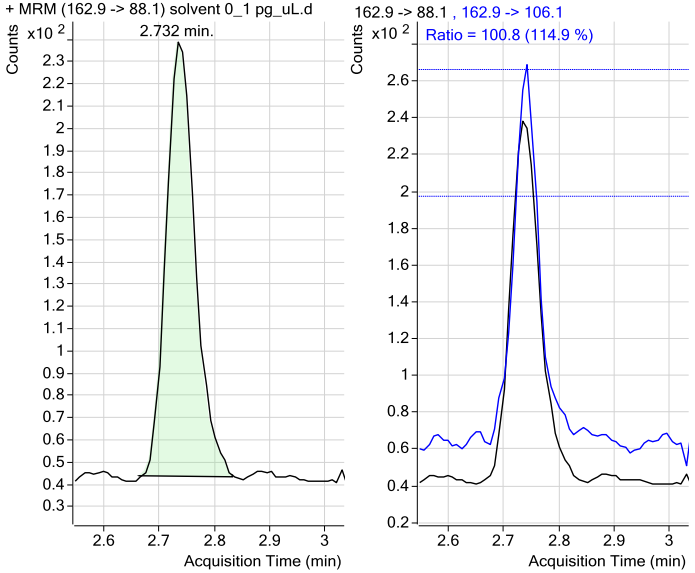
Recoveries for dilution in black tea

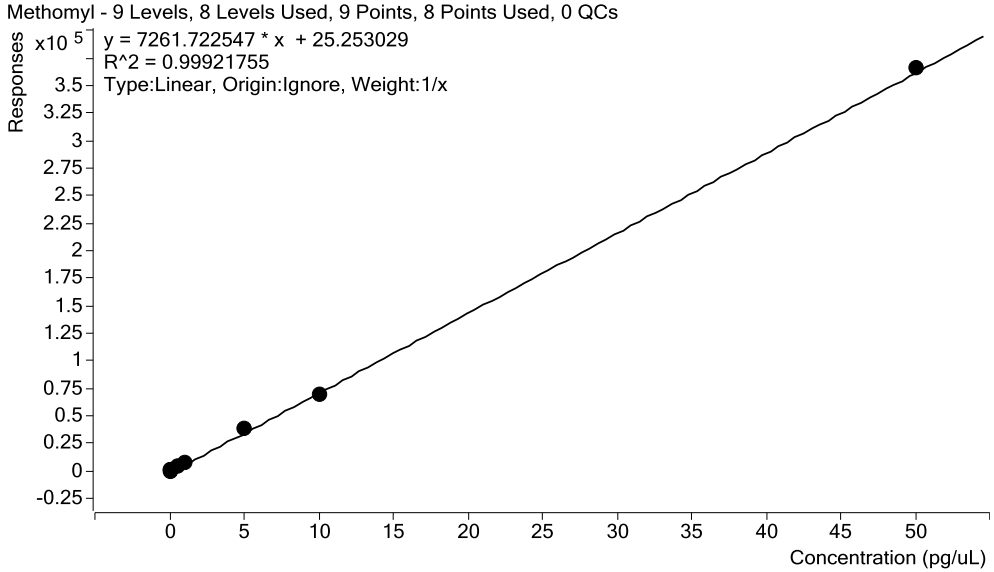


Matrix	Concentration	Recovery
Black tea	0.25	24.6
Black tea 1 in 2	0.22	44.9
Black tea 1 in 5	0.14	72.2
Black tea 1 in 10	0.078	77.6

Results for Methomyl

Calibration in solvent and recoveries in different matrices

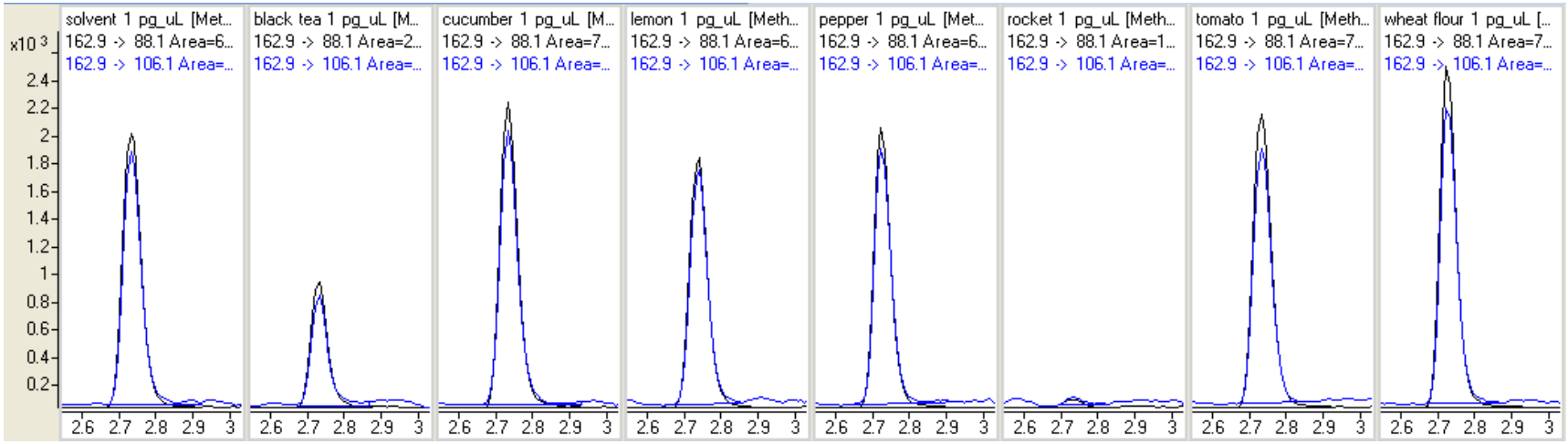




- Quantifier: 162.9 → 88.1
- Qualifier: 162.9 → 106.1
- Peak width: 12.6 s
- Cycle time: 600 ms
- Data points: 21

Results for Methomyl

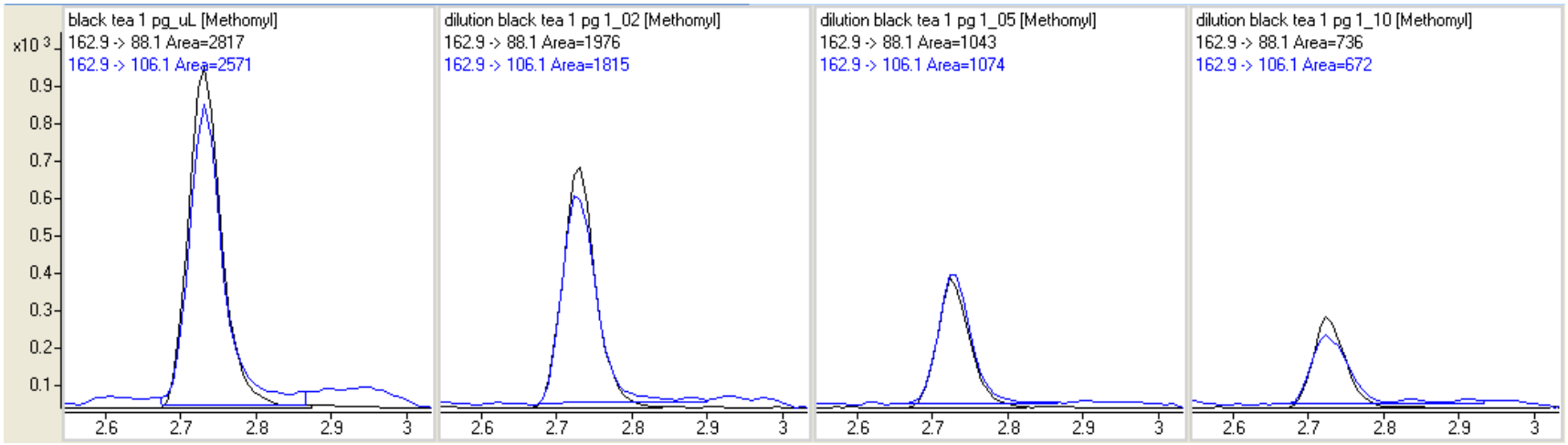
Calibration in solvent and recoveries in different matrices



Matrix	Concentration	Recovery
Solvent	0.94	
Black tea	0.38	38.5
Cucumber	1.00	99.8
Lemon	0.83	83.0
Pepper	0.85	85.5
Rocket	0.022	2.2
Tomato	0.99	98.9
Wheat flower	103.3	103.3

Results for Methomyl

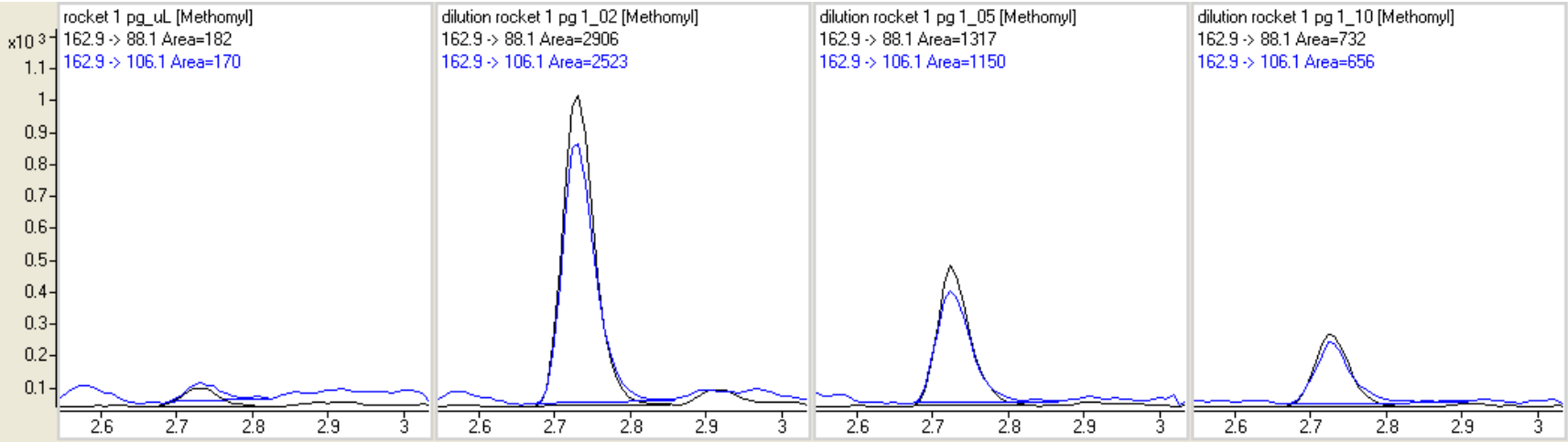
Recoveries for dilution in black tea



Matrix	Concentration	Recovery
Black tea	0.38	38.5
Black tea 1 in 2	0.354	53.7
Black tea 1 in 5	0.175	70.1
Black tea 1 in 10	0.088	97.8

Results for Methomyl

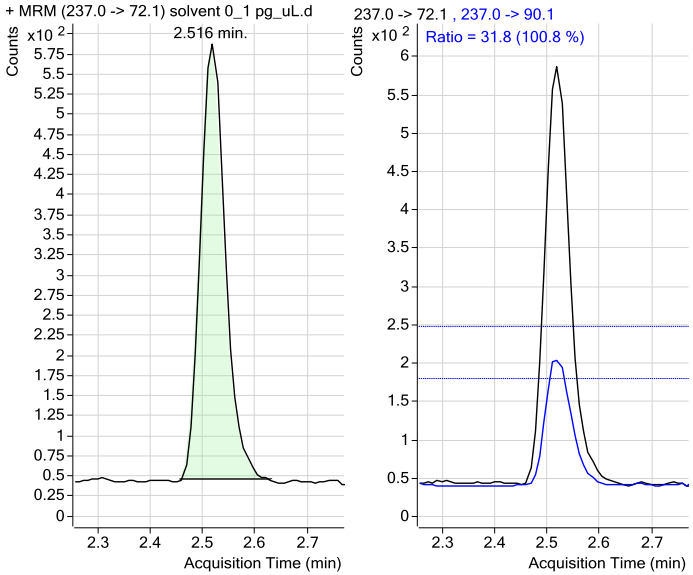
Recoveries for dilution in rocket

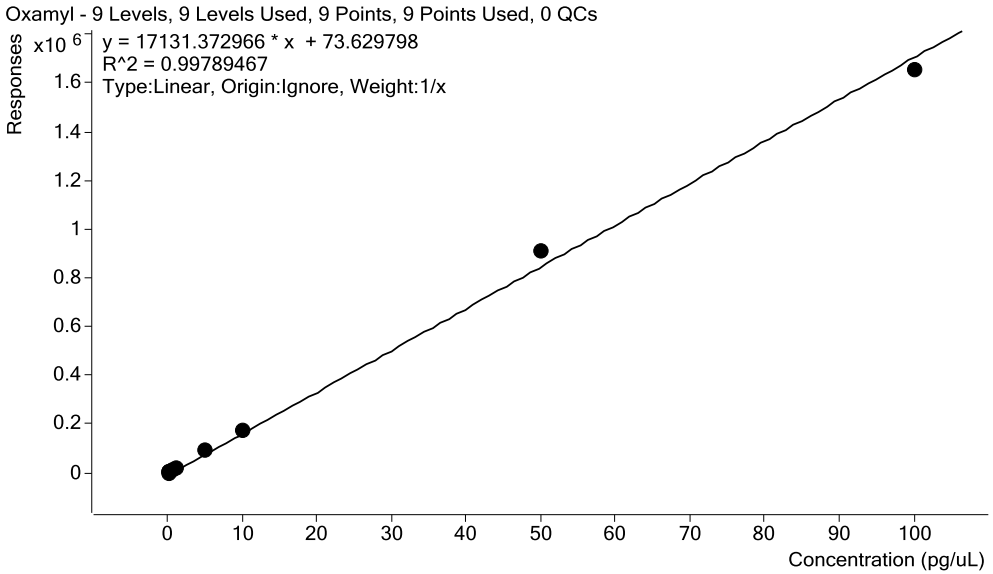


Matrix	Concentration	Recovery
Rocket	0.022	2.2
Rocket 1 in 2	0.397	79.3
Rocket 1 in 5	0.178	89.0
Rocket 1 in 10	0.0973	97.3

Results for Oxamyl

Calibration in solvent and recoveries in different matrices

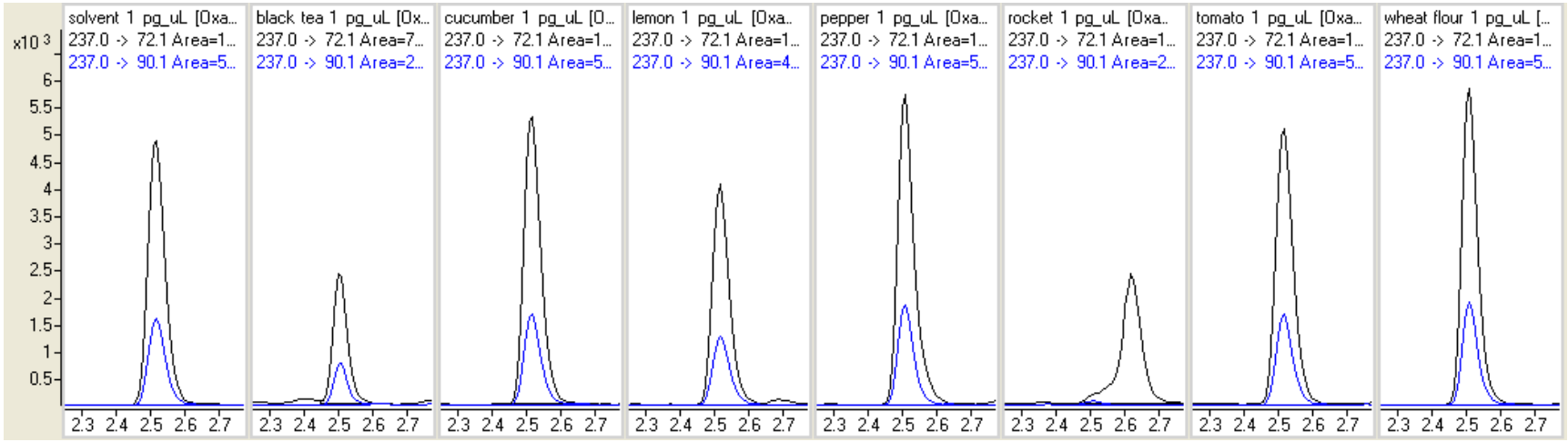




- Quantifier: 237.0 → 72.1
- Qualifier: 237.0 → 90.1
- Peak width: 12.6 s
- Cycle time: 600 ms
- Data points: 21

Results for Oxamyl

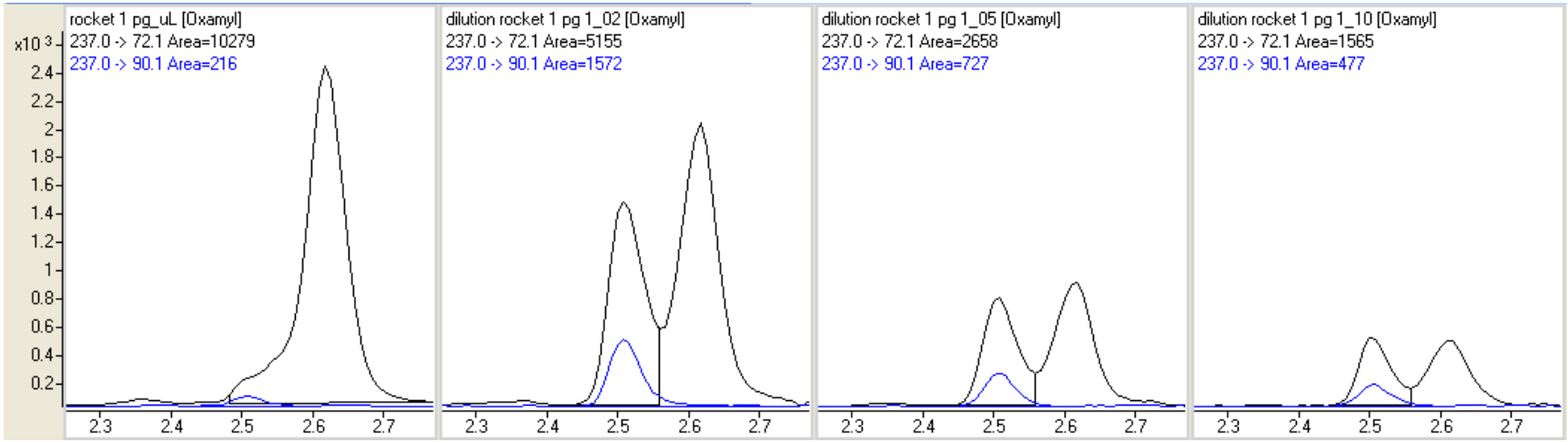
Calibration in solvent and recoveries in different matrices



Matrix	Concentration	Recovery
Solvent	0.98	
Black tea	0.41	40.6
Cucumber	1.05	105.1
Lemon	0.79	78.8
Pepper	1.10	109.6
Rocket	0.60	59.6
Tomato	1.03	103.3
Wheat flower	1.06	106.0

Results for Oxamyl

Recoveries for dilution in rocket



Matrix	Concentration	Recovery
Rocket	0.035**	3.5
Rocket 1 in 2	0.297	59.3
Rocket 1 in 5	0.151	75.4
Rocket 1 in 10	0.087	87.0

Why do we have triggered MRM?

Quantitation with Confirmation: Fingerprinting

<u>Full Scan Approach:</u>
Scan the entire fingerprint

<u>tMRM Approach:</u>
Focus on known fingerprint features

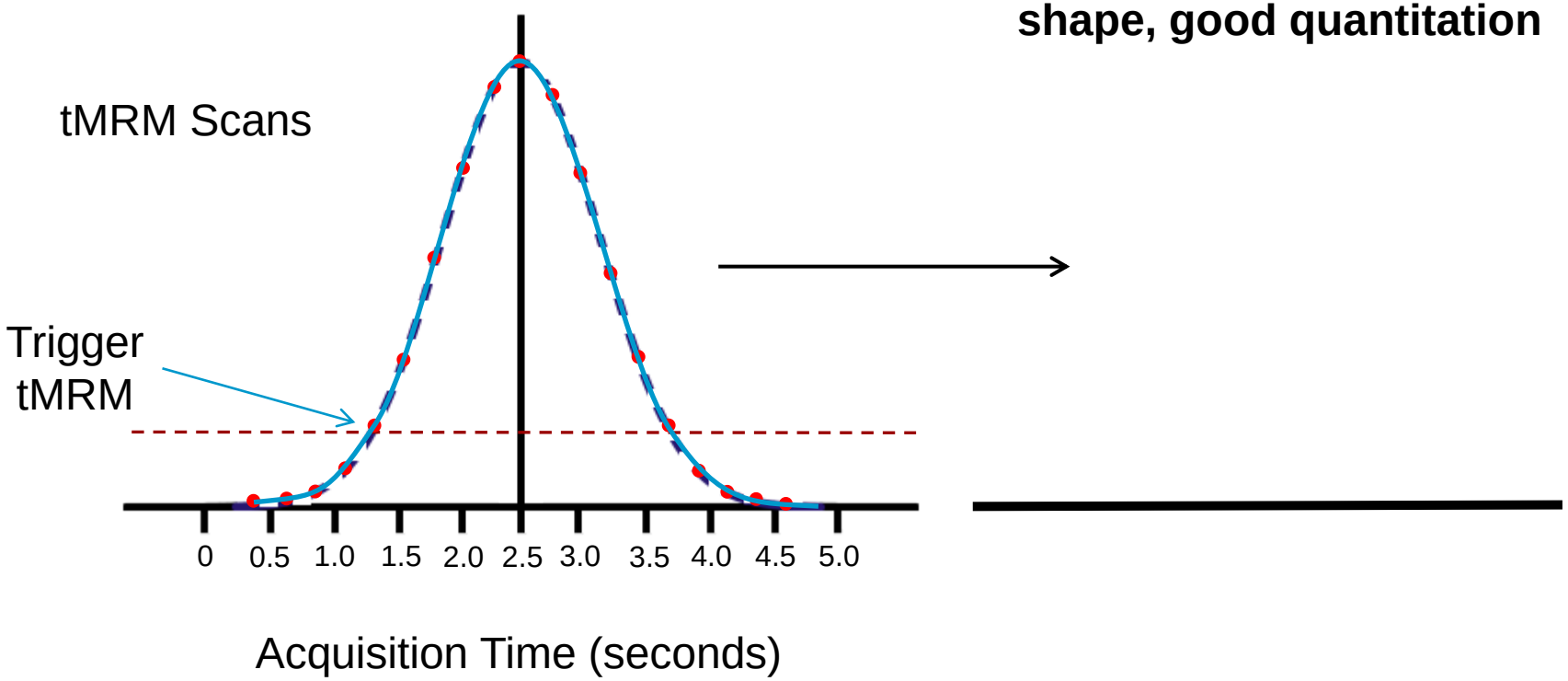
<u>Two possible scenarios:</u>

- Confirmation of positive findings with additional information (spectral matching)
- Elimination of potential false detects caused by matrix interferences

tMRM Scanning

tMRM Scan ≤ 50ms

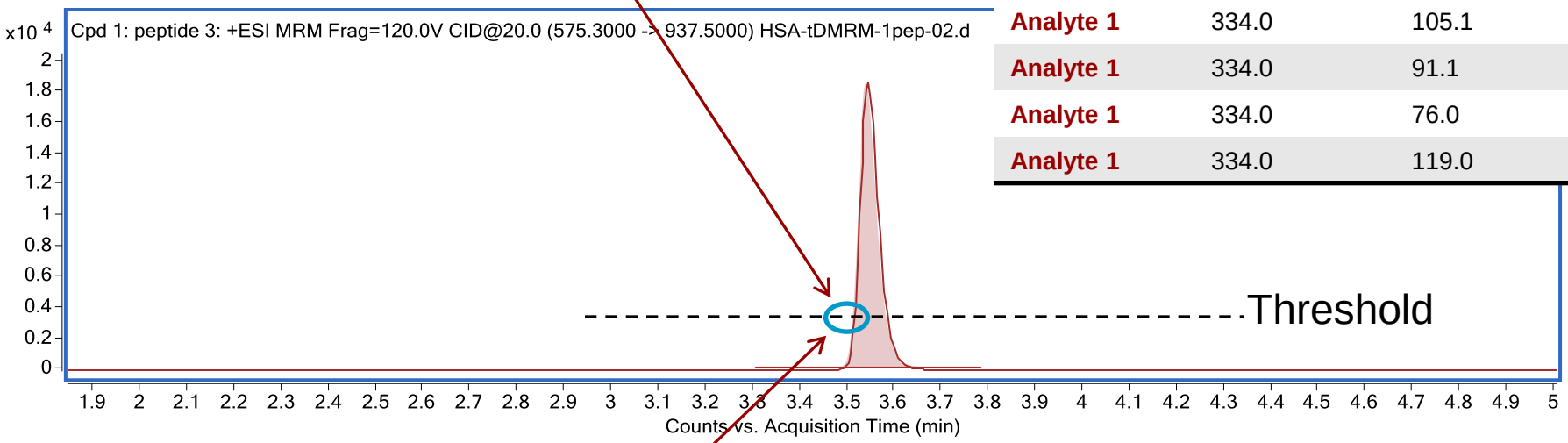
UHPLC compatible, good peak shape, good quantitation



Ideal situation: 1 Peak 5 sec wide, no Matrix

Triggered MRM (tMRM) Analysis

Secondary MRM
Transitions are “Triggered”



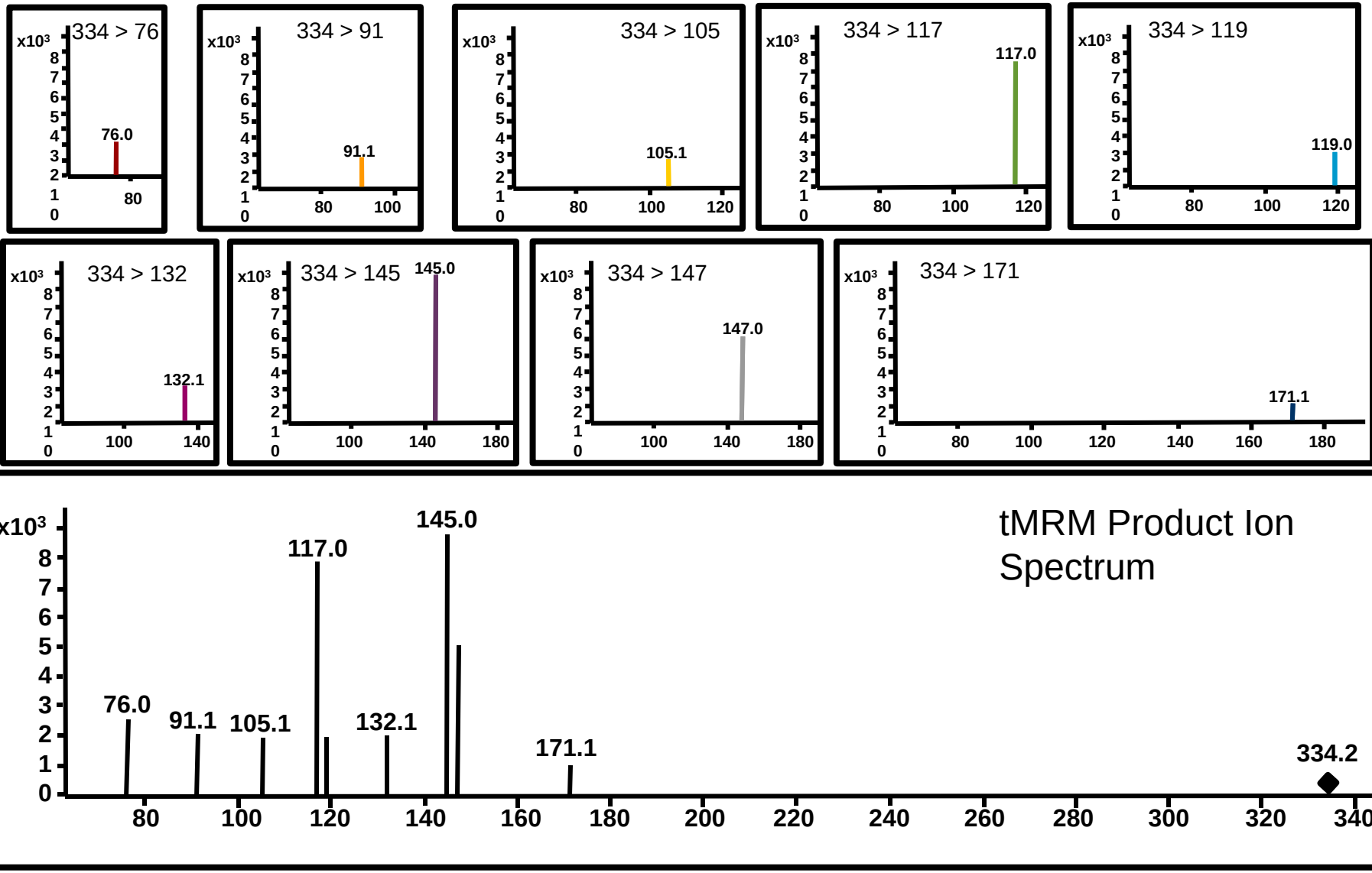
Triggered cycle (above threshold)

Compound	Precursor	Product
Analyte 1	334.0	145.0
Analyte 1	334.0	117.0
Analyte 1	334.0	132.1
Analyte 1	334.0	105.1
Analyte 1	334.0	91.1
Analyte 1	334.0	76.0
Analyte 1	334.0	119.0

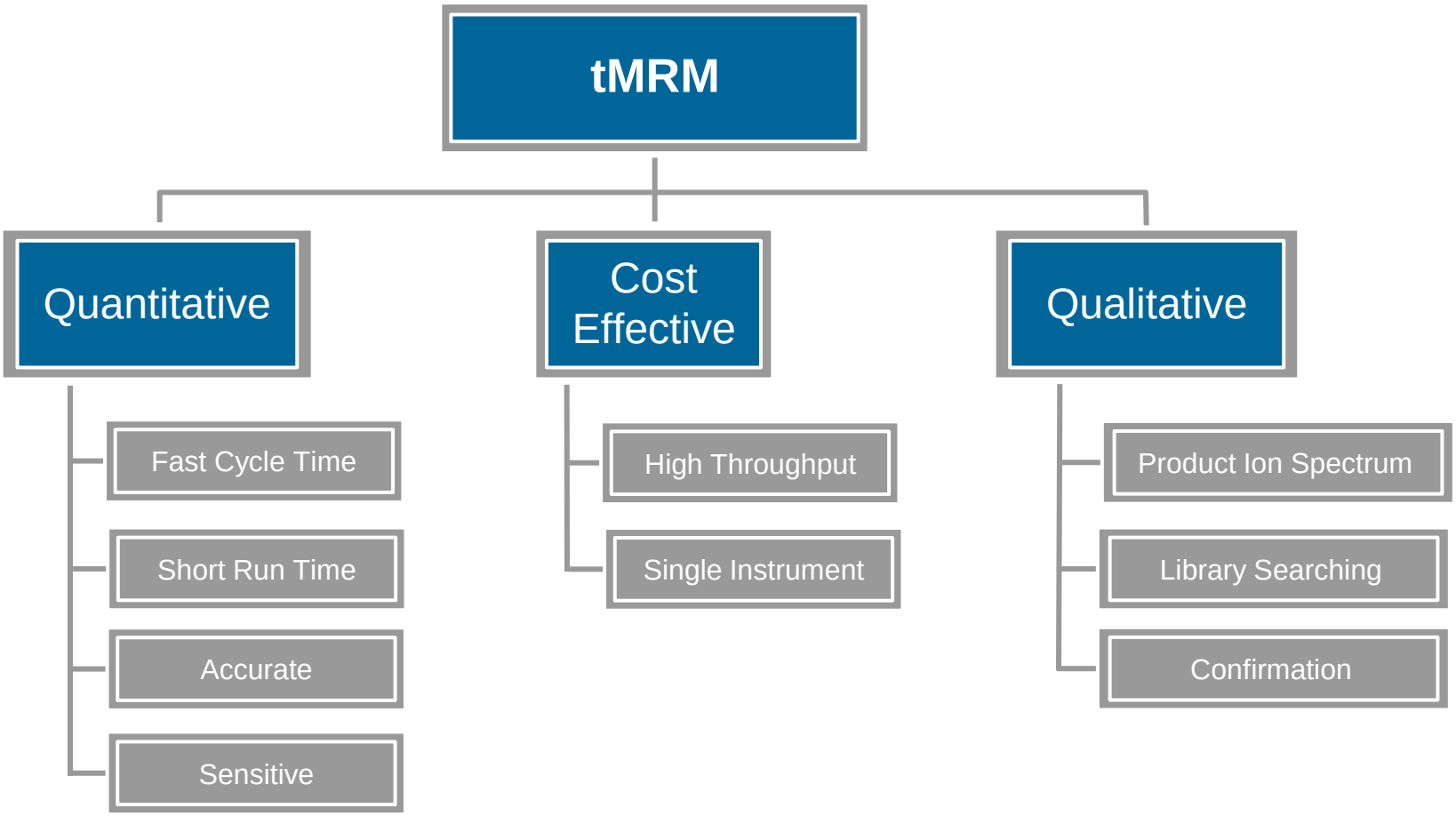
Primary cycle (below threshold)

Compound	Precursor	Product
Analyte 1	334.0	145.0
Analyte 1	334.0	117.0

tMRM Product Ion Spectrum



The Ideal Analytical Solution



tMRM LC/MS Application Kits

Targeted Screening with QQQ

Pesticides

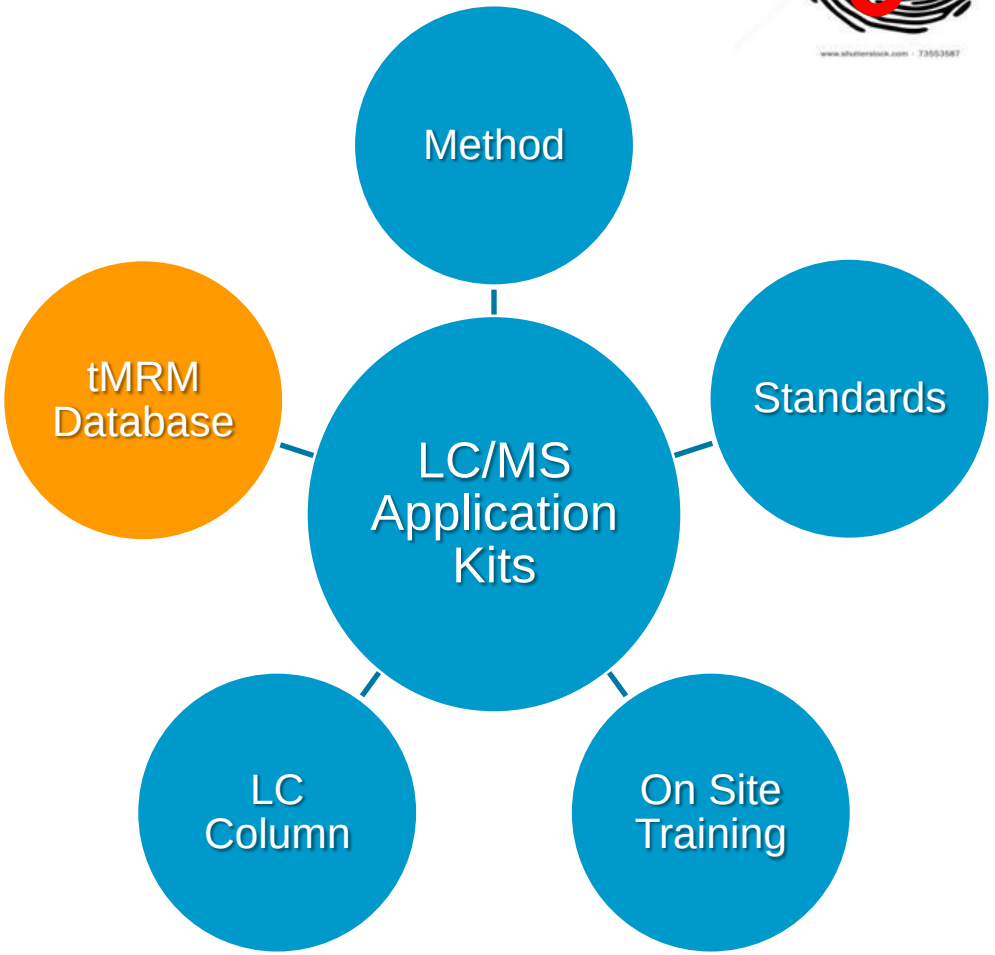
-Test Mix: 250+ compounds
-DB: 500+ compounds
-Library: 250+ compounds

Veterinary Drugs

-Test Mix: 200+ compounds
-DB: 500+ compounds
-Library: 200+ compounds

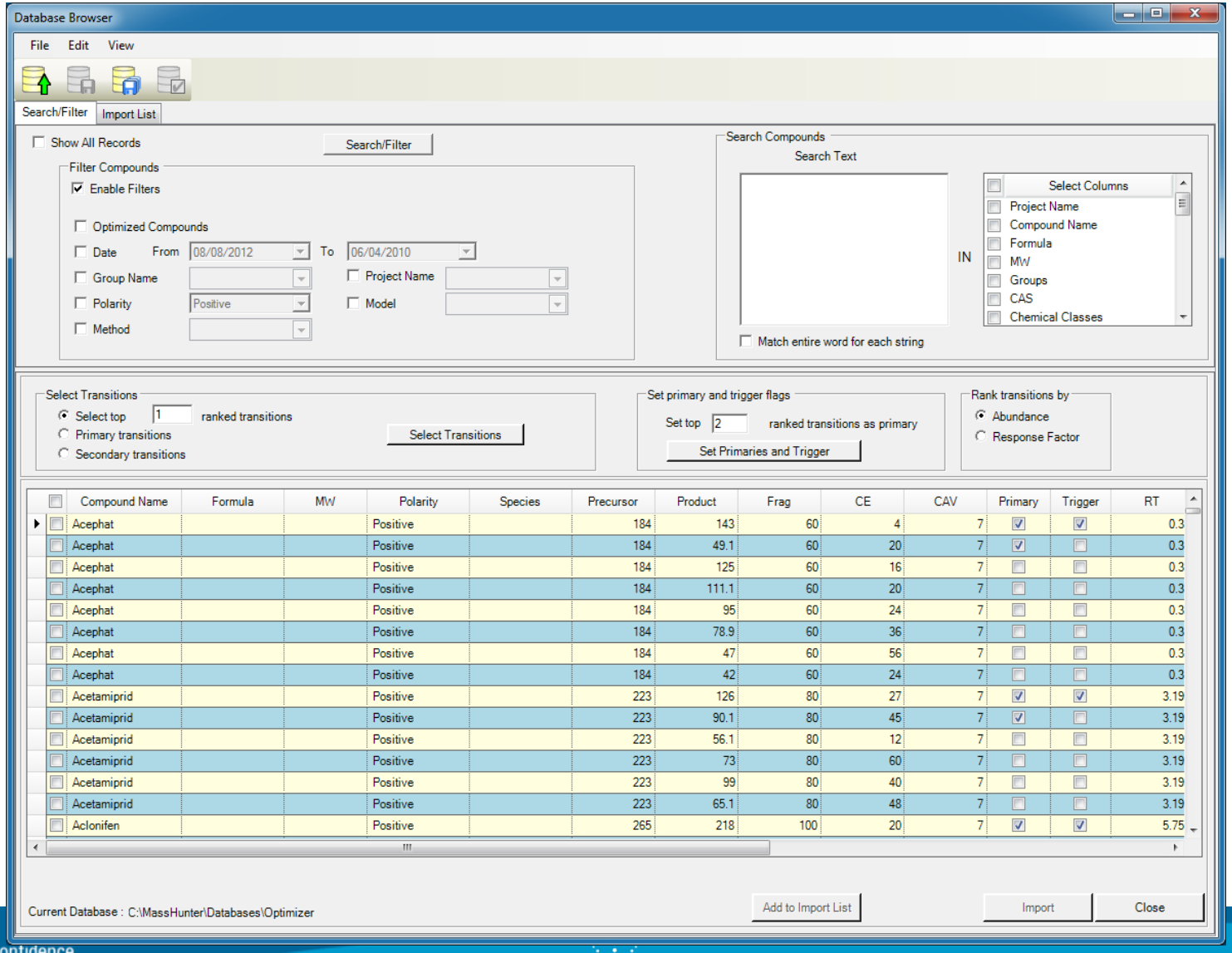
Forensic Toxicology

-Test Mix: 100+ compounds
-DB: 500+ compounds
-Library: 100+ compounds



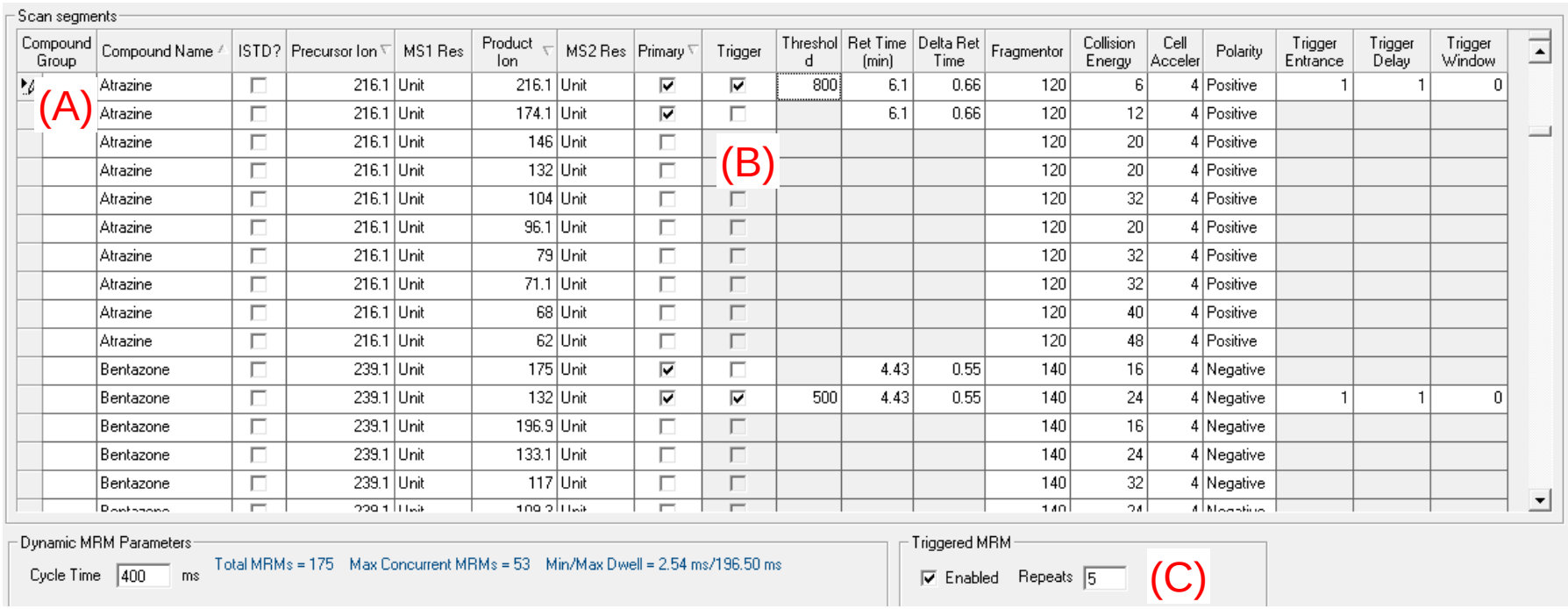
Triggered MRM database and library

Optimizer database



Triggered MRM Parameters

Primary transitions (A) with defined threshold (B) trigger additional transitions for given number of repeats (C).



Scan segments

Compound Group	Compound Name	ISTD?	Precursor Ion	MS1 Res	Product Ion	MS2 Res	Primary	Trigger	Threshold	Ret Time (min)	Delta Ret Time	Fragmentor	Collision Energy	Cell Acceler	Polarity	Trigger Entrance	Trigger Delay	Trigger Window
(A)	Atrazine	☐	216.1	Unit	216.1	Unit	☑	☑	800	6.1	0.66	120	6	4	Positive	1	1	0
	Atrazine	☐	216.1	Unit	174.1	Unit	☑	☐		6.1	0.66	120	12	4	Positive			
	Atrazine	☐	216.1	Unit	146	Unit	☐					120	20	4	Positive			
	Atrazine	☐	216.1	Unit	132	Unit	☐	(B)				120	20	4	Positive			
	Atrazine	☐	216.1	Unit	104	Unit	☐	☐				120	32	4	Positive			
	Atrazine	☐	216.1	Unit	96.1	Unit	☐	☐				120	20	4	Positive			
	Atrazine	☐	216.1	Unit	79	Unit	☐	☐				120	32	4	Positive			
	Atrazine	☐	216.1	Unit	71.1	Unit	☐	☐				120	32	4	Positive			
	Atrazine	☐	216.1	Unit	68	Unit	☐	☐				120	40	4	Positive			
	Atrazine	☐	216.1	Unit	62	Unit	☐	☐				120	48	4	Positive			
	Bentazone	☐	239.1	Unit	175	Unit	☑	☐		4.43	0.55	140	16	4	Negative			
	Bentazone	☐	239.1	Unit	132	Unit	☑	☑	500	4.43	0.55	140	24	4	Negative	1	1	0
	Bentazone	☐	239.1	Unit	196.9	Unit	☐	☐				140	16	4	Negative			
	Bentazone	☐	239.1	Unit	133.1	Unit	☐	☐				140	24	4	Negative			
	Bentazone	☐	239.1	Unit	117	Unit	☐	☐				140	32	4	Negative			

Dynamic MRM Parameters

Cycle Time 400 ms Total MRMs = 175 Max Concurrent MRMs = 53 Min/Max Dwell = 2.54 ms/196.50 ms

Triggered MRM

☑ Enabled Repeats 5 (C)

How does triggered MRM compare with traditional MRM methods

- Method was setup with more than 120 pesticides
- In tMRM acquistion was done with 2 primary transitions
- Triggering happened above a set threshold set for every compound
- In comparison an traditional dynamic MRM was setup with the same 2 transitions to compare the quantitative performance of the method
- Analysis of real samples to test tMRM

Method parameters

UHPLC method

UHPLC column	Agilent ZORBAX Eclipse Plus C18 RRHD 2.1 x 150 mm, 1.8 µm @ 30°C
Mobile phase	A: 5 mM NH_4 formate + 0.1% formic acid B: 5 mM NH_4 formate + 0.1% formic acid in methanol
Gradient program	Min % B 0 5 0.5 5 3.0 40 17 100 19 100 19.1 5 Stop time 22 min
Flow rate	0.40 ml/min
Injection volume	3 µl

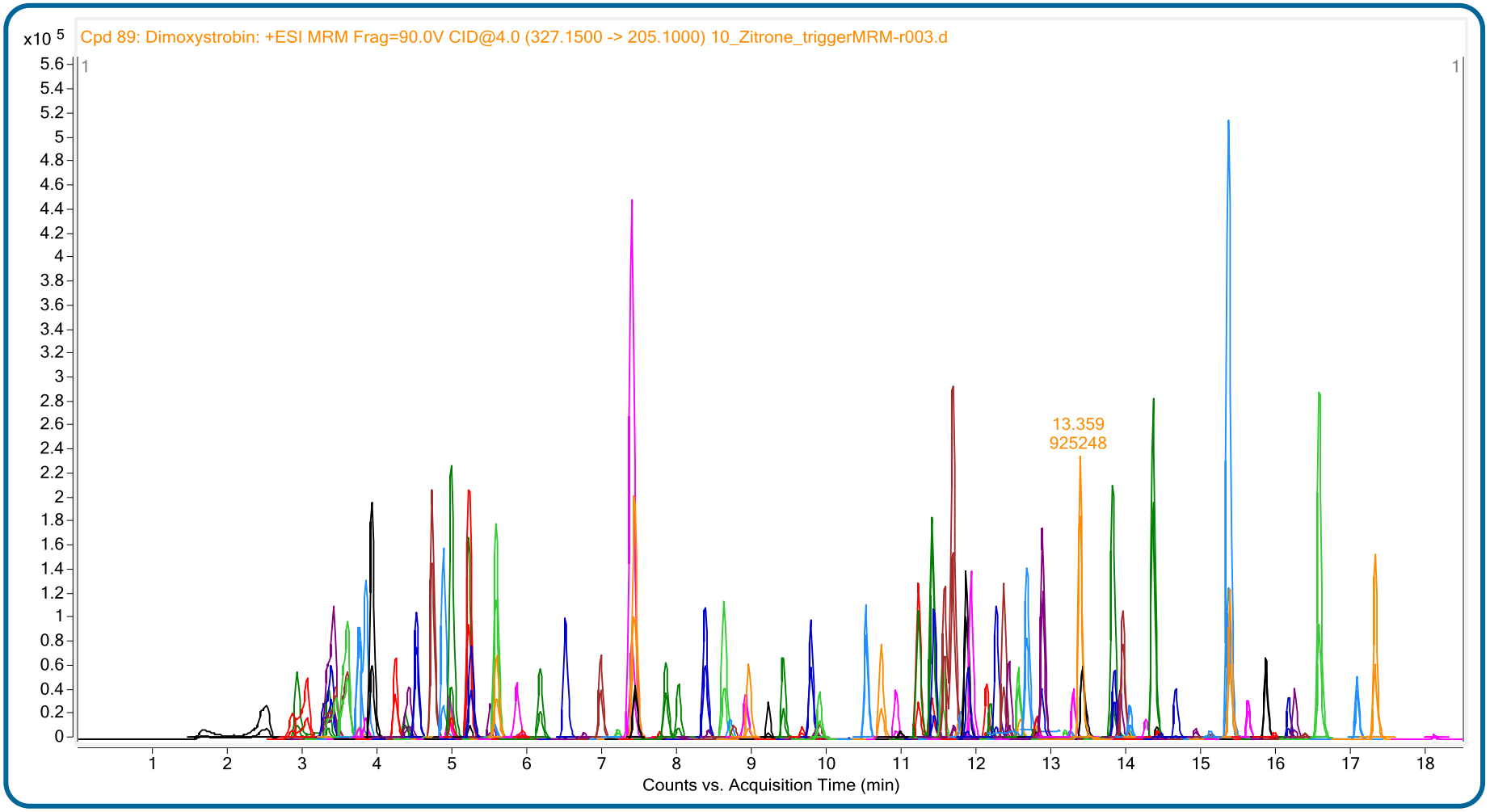
MS method

Electrospray ionization

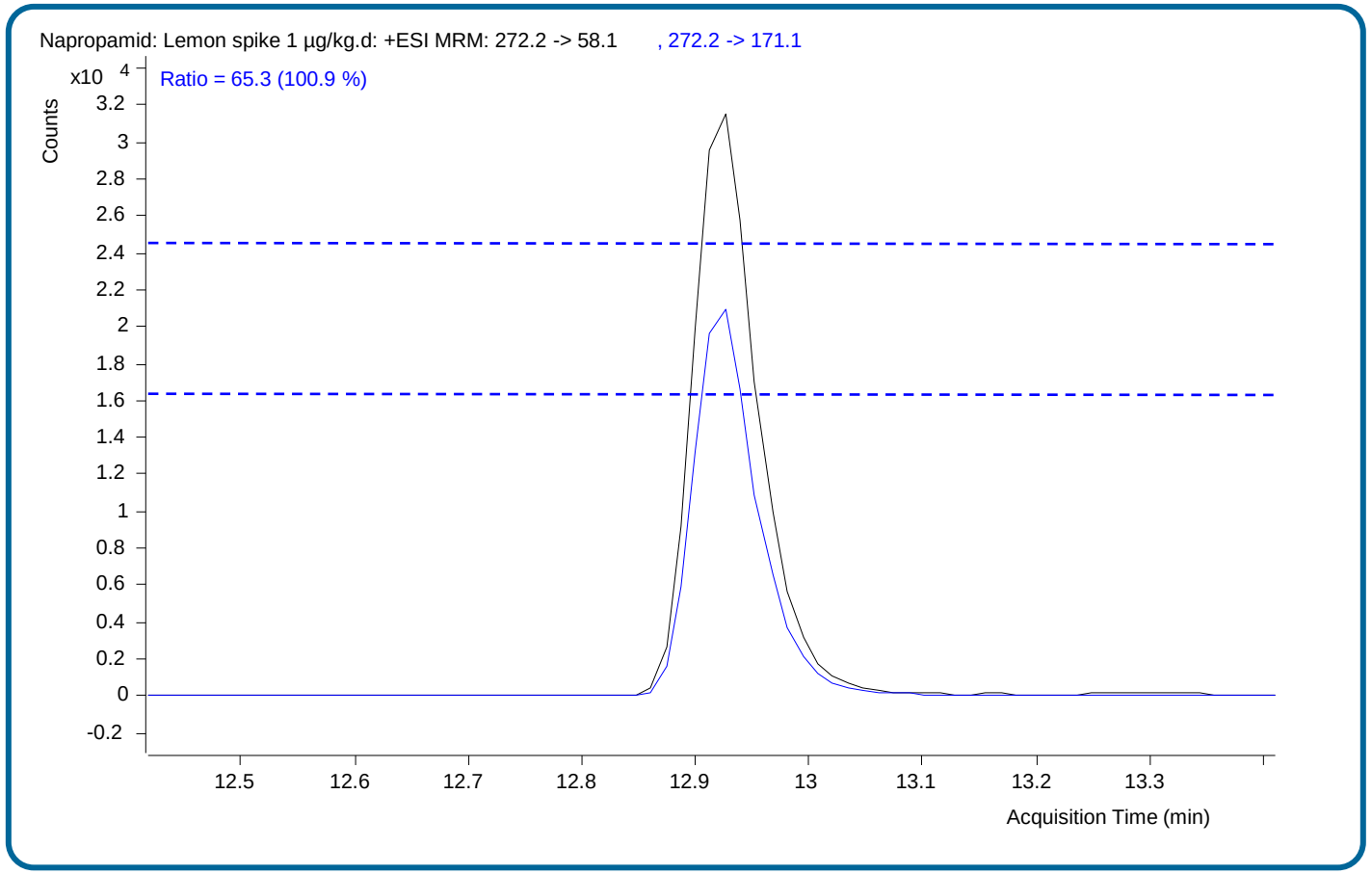
Triggered MRM: 2 primary transitions and up to 8 additional confirmatory ions

Dynamic MRM: 2 transitions per compound

UHPLC-MS/MS chromatogram at 10ng/mL in lemon extract

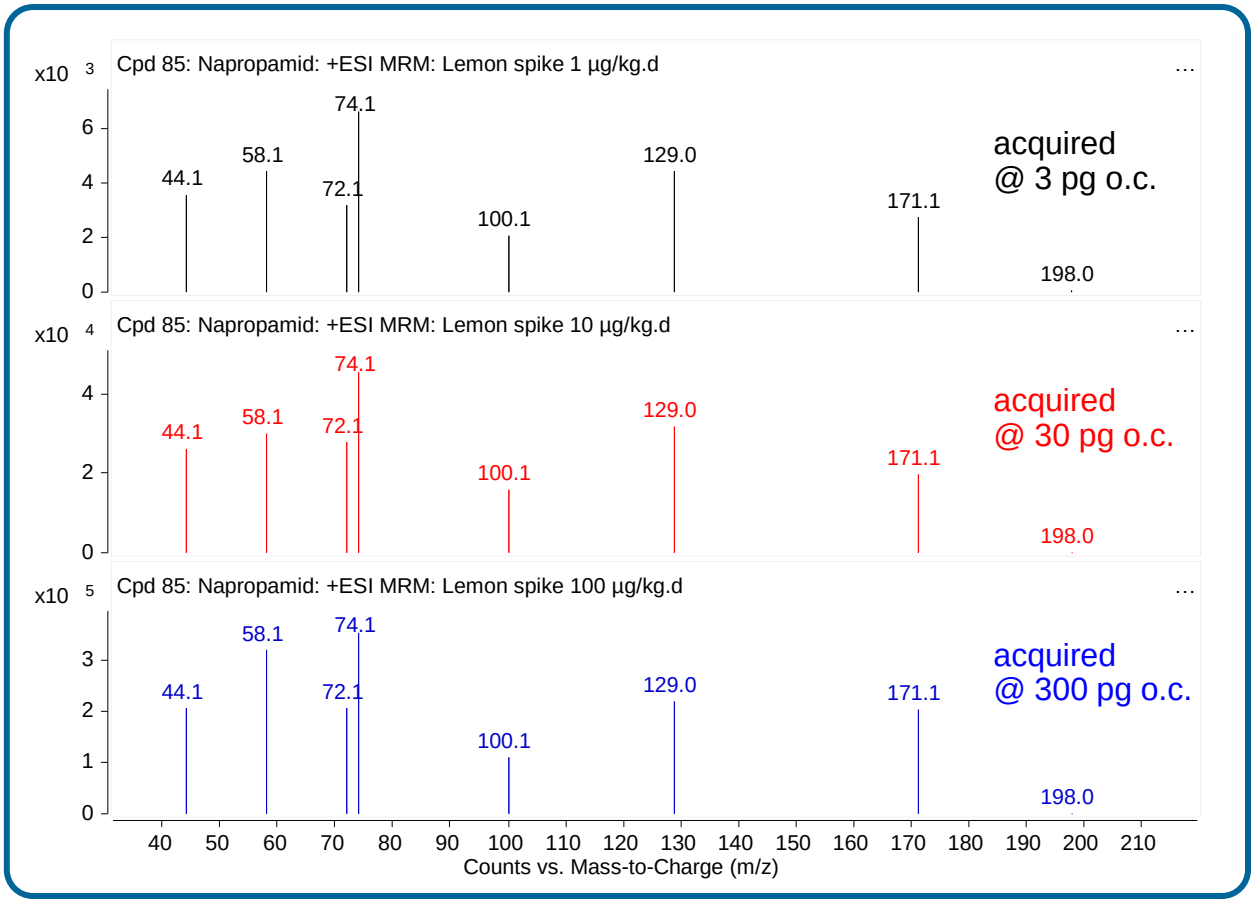


Real samples – Napropamide in Lemon extract



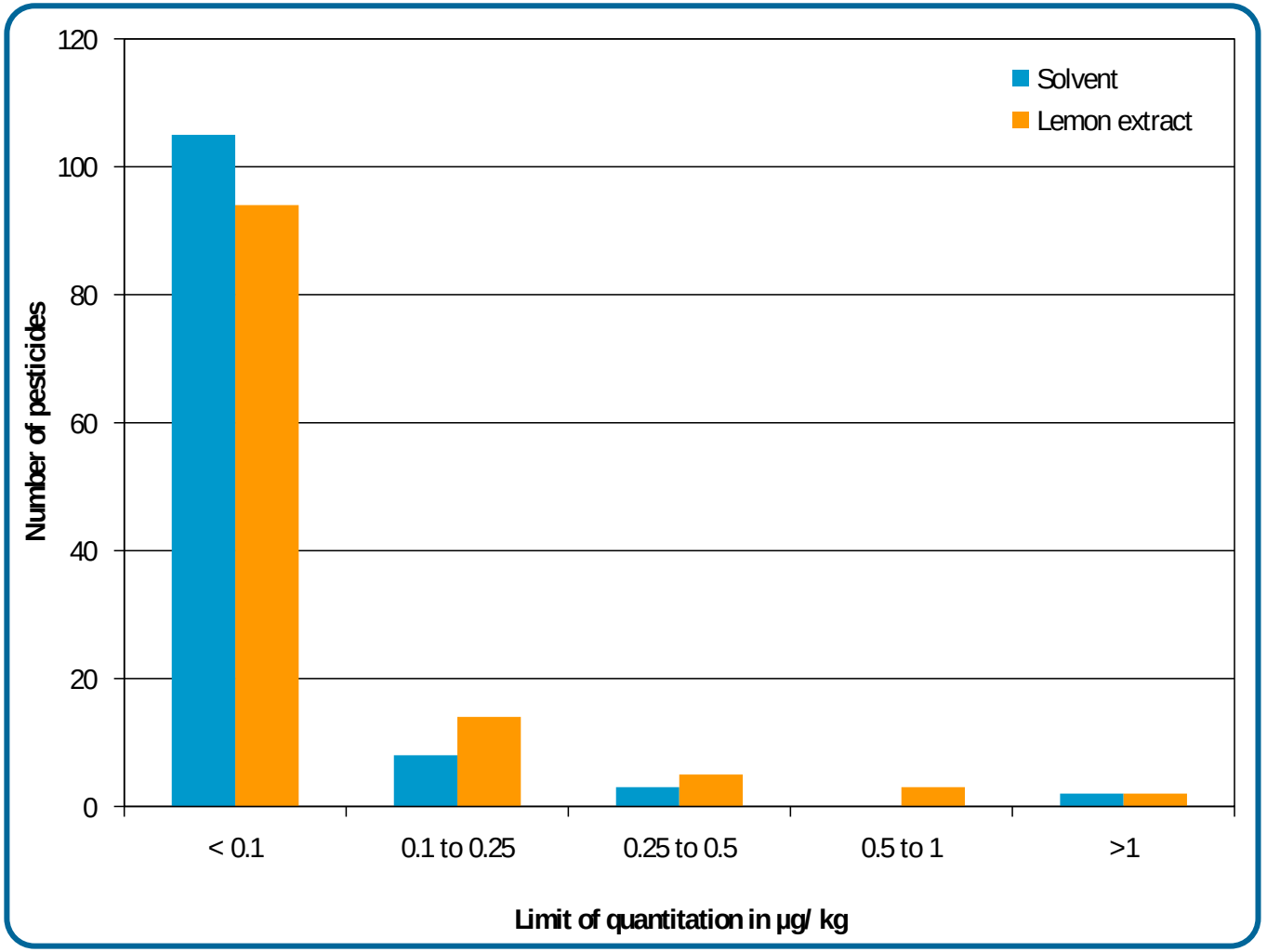
MRM chromatogram of the two primary transitiitions spiked at 1µg/kg

Real samples – tMRM spectra for Napropamide in Lemon at different concentrations

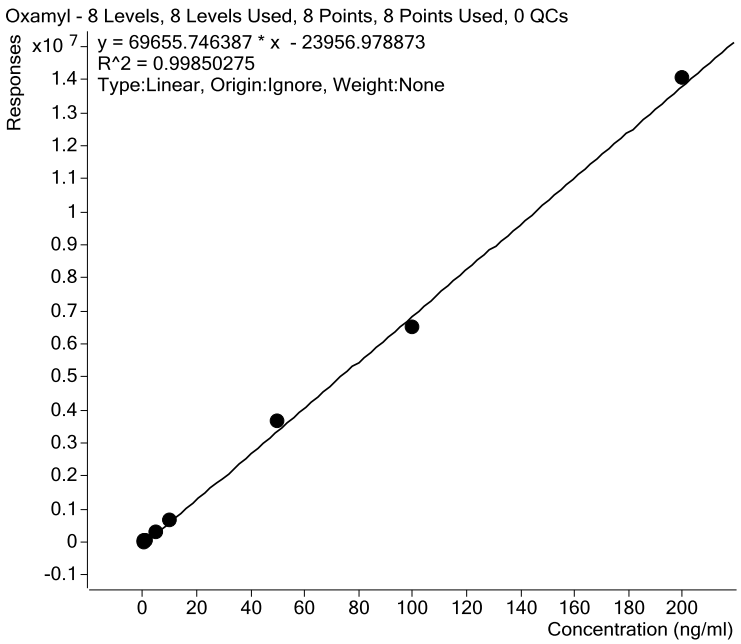


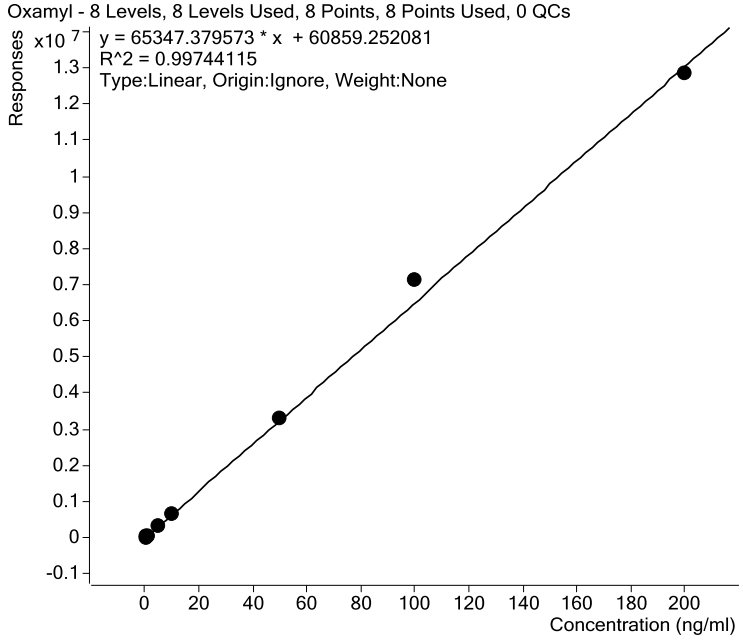
tMRM spectra at 1µg/kg, 10µg/kg and 100 µg/kg

Method validation – Limits of quantitation



Quantitative performance Oxamyl

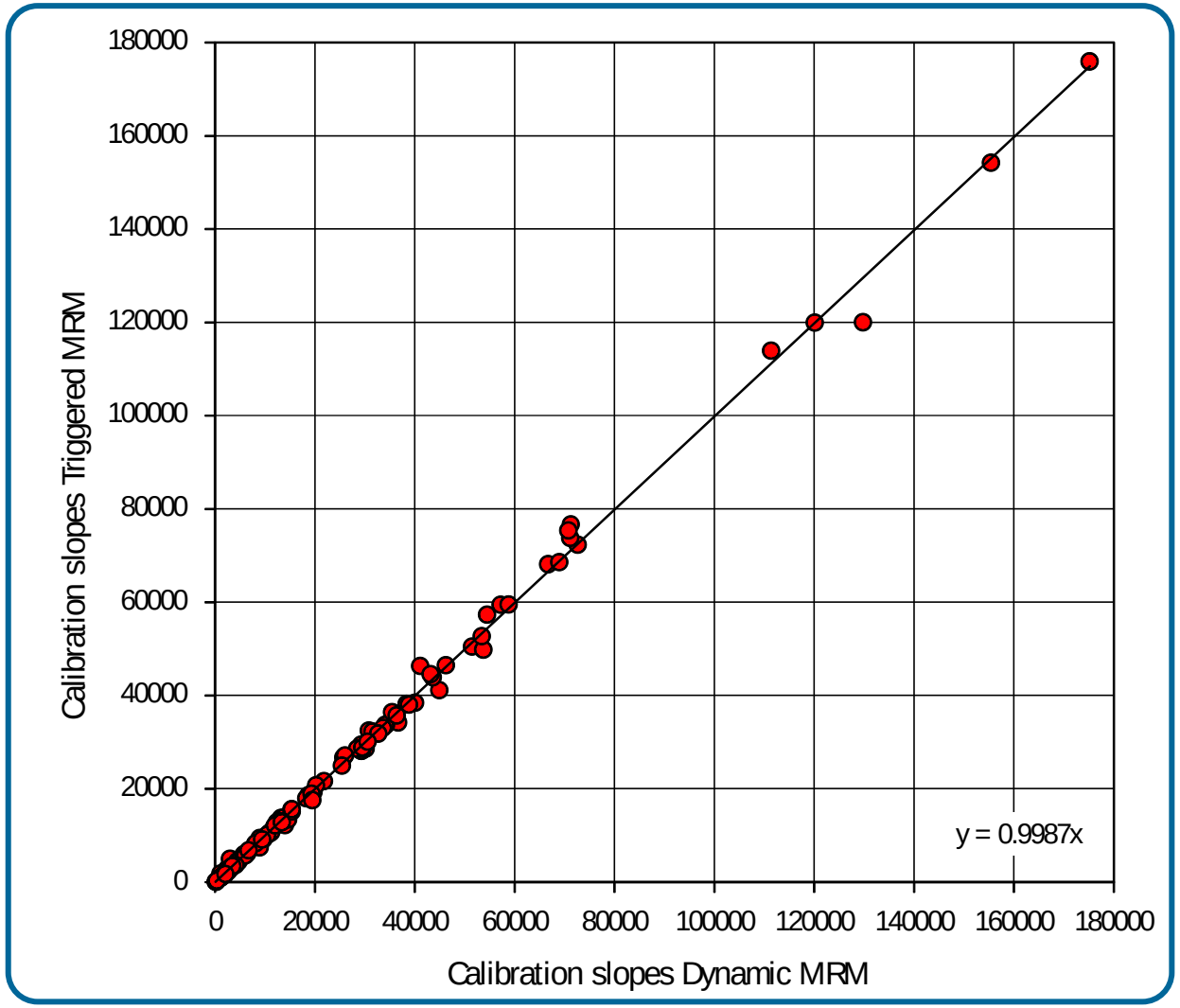




dMRM calibration curve

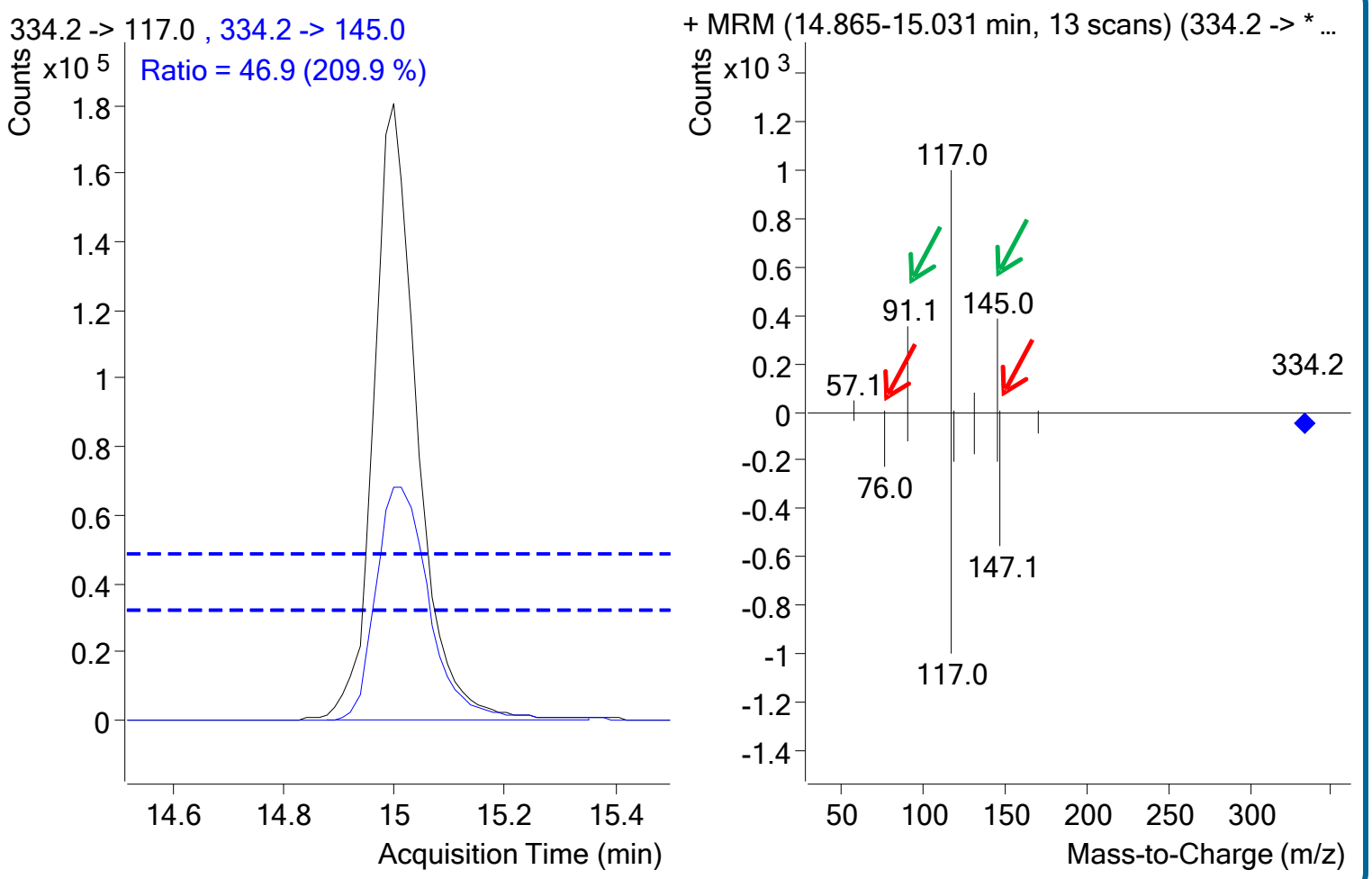
tMRM calibration curve

Quantitative performance

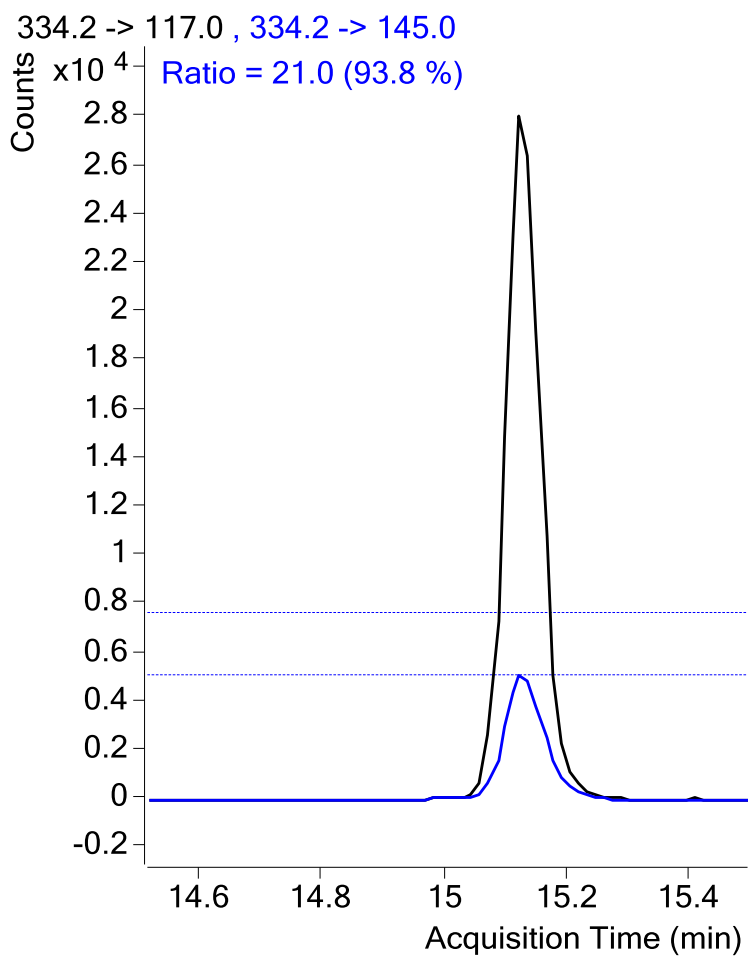


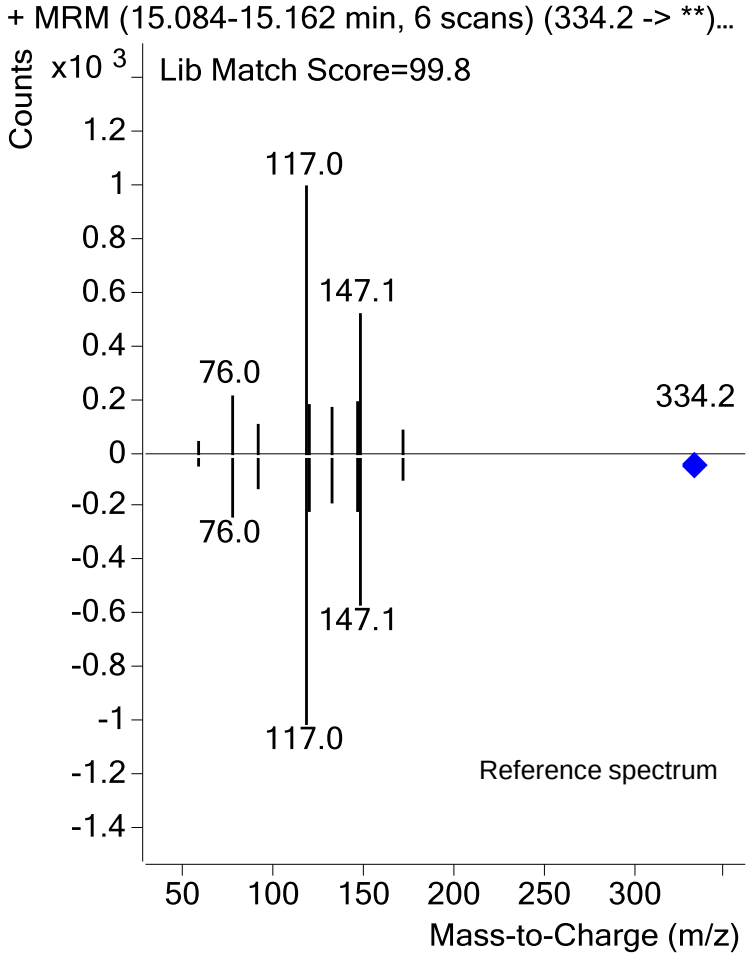
Calibration curve slopes of all pesticides in the method

Real samples – Tebufenpyrad in ginger extract

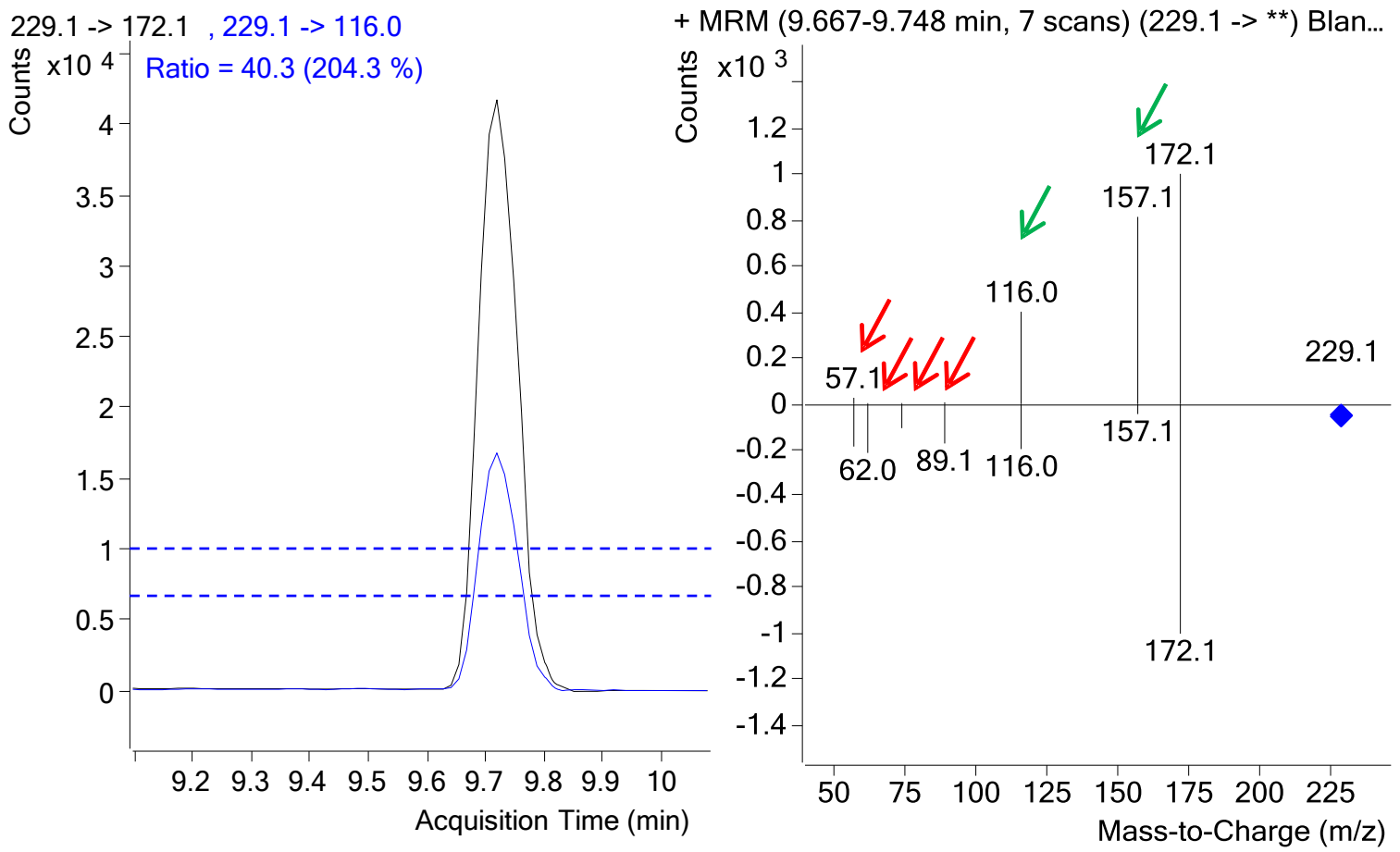


Real samples – Tebufenpyrad in ginger extract

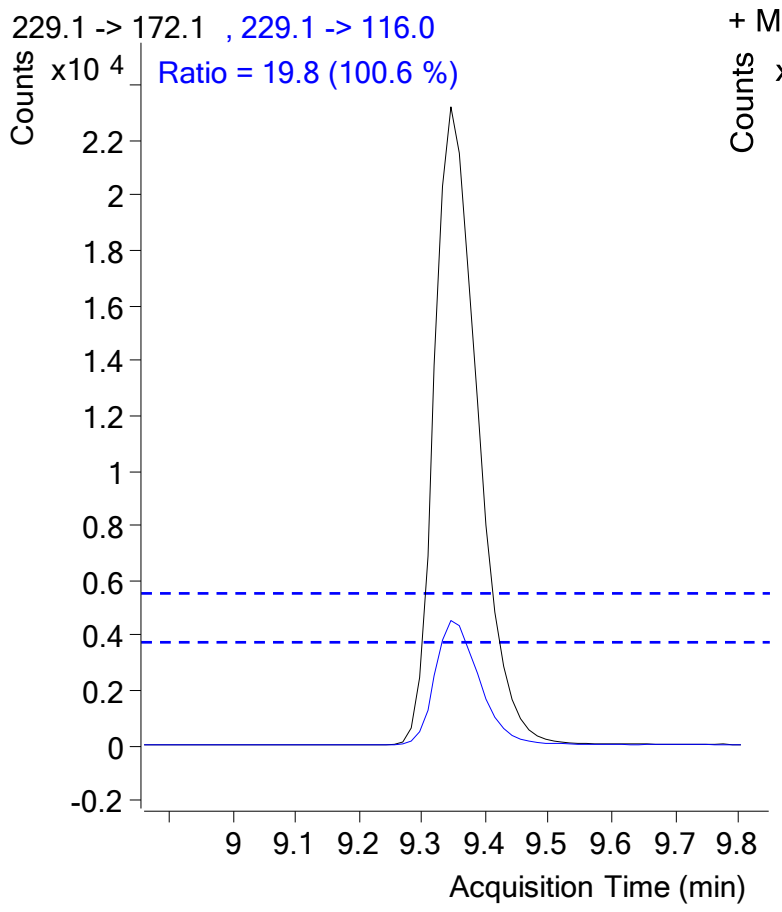


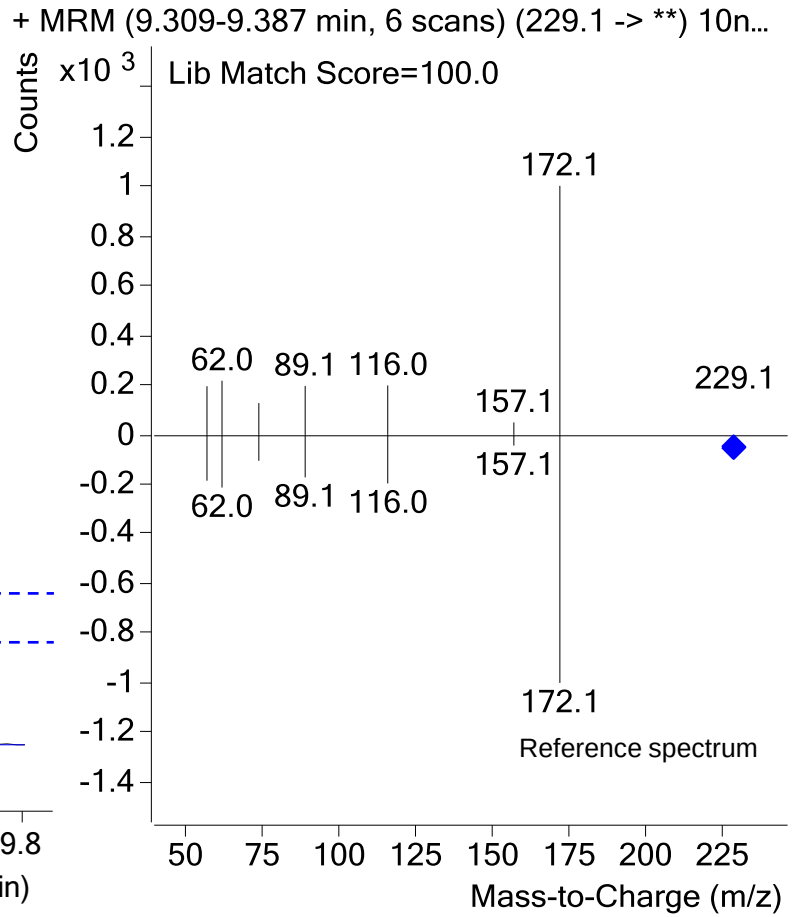


Real samples – Tebuthiuron in chamomile extract



Real samples – Tebuthiuron in chamomile extract



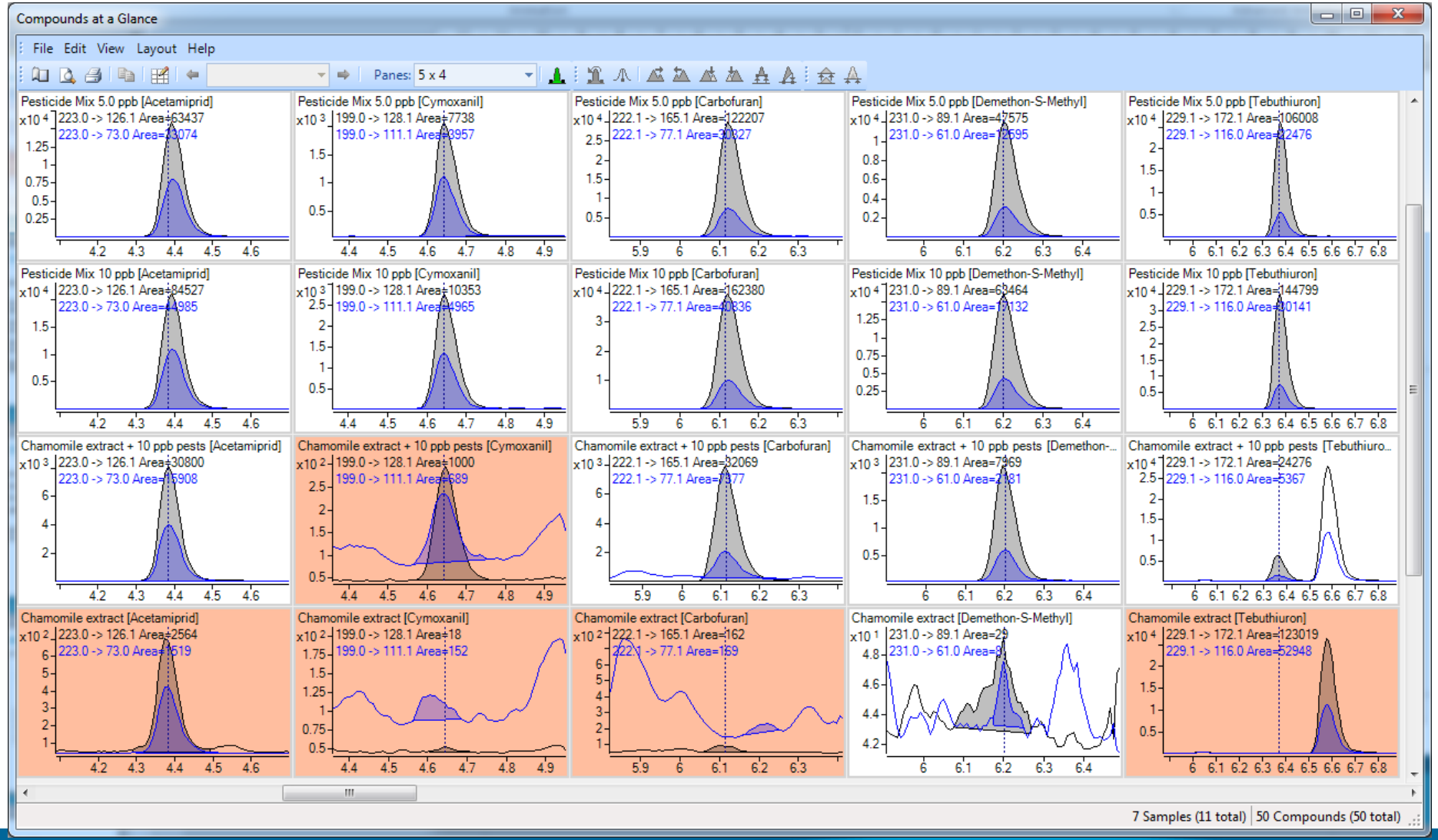


Real samples – examples for matrix interferences

			Library match score	
Pesticide	**Matrix**		**Target compound**	**Matrix interference**
Dichlovos	Lemon		94.5 %	78.1 %
Thifensulfuron-methyl	Green Tea		96.6 %	71.5 %
Tebufenpyrad	Ginger		99.8 %	55.9 %
Tebuthiuron	Chamomile		97.8 %	58.0 %
Imazalil	Chamomile		99.8 %	58.1 %
Terbutylazin	Chamomile		99.6 %	82.1 %

Compounds at a glance

Efficient review of results using outlier flagging



Summary and conclusions (1)

- Combination of 1290 UHPLC with the G6490 QQQ allows for short run times and large pesticide lists
- Validation resulted in excellent precision and low limits of quantitation for well over 300 priority pesticides even in challenging matrix types
- Dilution is a possible way to overcome matrix effects like ion suppression and often allows for the quantitation based on a solvent calibration
- With any dilution an improved robustness of the method can be expected due to the lower matrix amount which is introduced into the ionization source
- Increasing numbers of analytes and challenging matrices increase the probability of erroneous signals on the MRM traces of pesticides potentially causing false detects

Summary and conclusions (2)

- Triggered MRM enables spectral confirmation at LOQ levels
- Same quantitative performance than traditional MRM methods
- Seamless integration with new triggered MRM databases and libraries
- Automatic reference library matching allows efficient data review and automatic flagging of suspect cases
- By using only one primary transition triggered MRM potentially extends the scope of multi-residue methods to up to twice as many compounds

Application notes for your reference

Triggered MRM: Simultaneous Quantitation and Confirmation Using Agilent Triple Quadrupole LC/MS Systems

Technical Overview

Triggered MRM (tMRM) acquisition is an analytical method which is available for all Agilent Triple Quadrupole LC/MS systems. tMRM acquisition combines MRM with the generation of a product ion spectrum which can then be used for library identification and confirmation. As a result, tMRM analysis decreases analysis time, increases throughput, and allows for fast, sensitive quantitative and qualitative analysis on a single instrument, in a single analytical run.

How tMRM Acquisition Works

In tMRM analysis, up to 10 MRM transitions (primary and secondary) are defined for each target analyte in the method. The primary transitions are acquired for all analytes, but when the signal of one of the primary transitions exceeds a user-defined threshold, the secondary transitions are "triggered" and acquired for a specified number of scans.

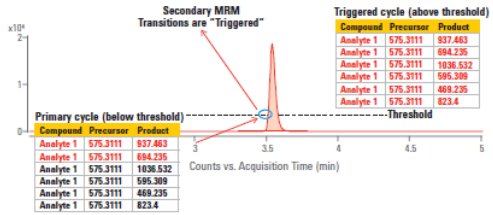


Figure 1. A tMRM experiment with two primary transitions for each analyte. Secondary MRM transitions are triggered when the primary MRM signals cross a user-defined threshold.

Agilent Technologies

5990-8461EN

Application of a Triggered MRM Database and Library for the Quantitation and Identification of Pesticides in Food Extracts

Application Note

Food

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Abstract

This application note describes the development of a triggered MRM database and library for more than 300 pesticides. It illustrates its use to analyze a range of food commodities with an LC/MS method developed for a specific suite of 120 pesticide residues. An Agilent 1290 Infinity LC System was coupled to an Agilent 6460 Triple Quadrupole LC/MS System and operated in positive electrospray using Agilent Jet Stream Technology. The triggered MRM acquisition mode was used for quantitation and verification and to eliminate potential false detects. A short, in-house, validation done for three commodity groups with five representative matrices showed that the developed triggered MRM method was appropriate for the analysis of pesticides in food extracts with regards to the required limits of quantitation (LOQs), linearity, and reproducibility. Several examples are shown where a high risk of an interfering matrix peak being incorrectly assigned as a pesticide, was mitigated through triggered MRM. Automatic reference library matching, displayed alongside quantitation results allows data to be reviewed efficiently and for suspect cases to be flagged automatically.

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