Current and future trends in Pesticide Residue analysis

Jim Garvey

- **A brief history of Pesticide residue analysis**

Contents

Where we have come from

- **1999**

GC

HPLC

ECD

PFPD

DAD

< 50 Pesticides mostly OC's and OP's

Carbendazim and Thiabendazole

Where we have come from

- Three analyses required for positive results – screen, confirm and quantify
- 12 different standard mixes to be made up and checked
- Very limited structural data
- High risk of false positives and false negatives

Why change ?

- Scope too limited to cover the pesticides being used and found in other countries
- EU pressure
- Residue definitions often include a significant number of metabolites
- Development of instruments with Mass Spectrometry detectors at affordable prices

Scope expansion

- **MS Techniques**

Full scan

SIM

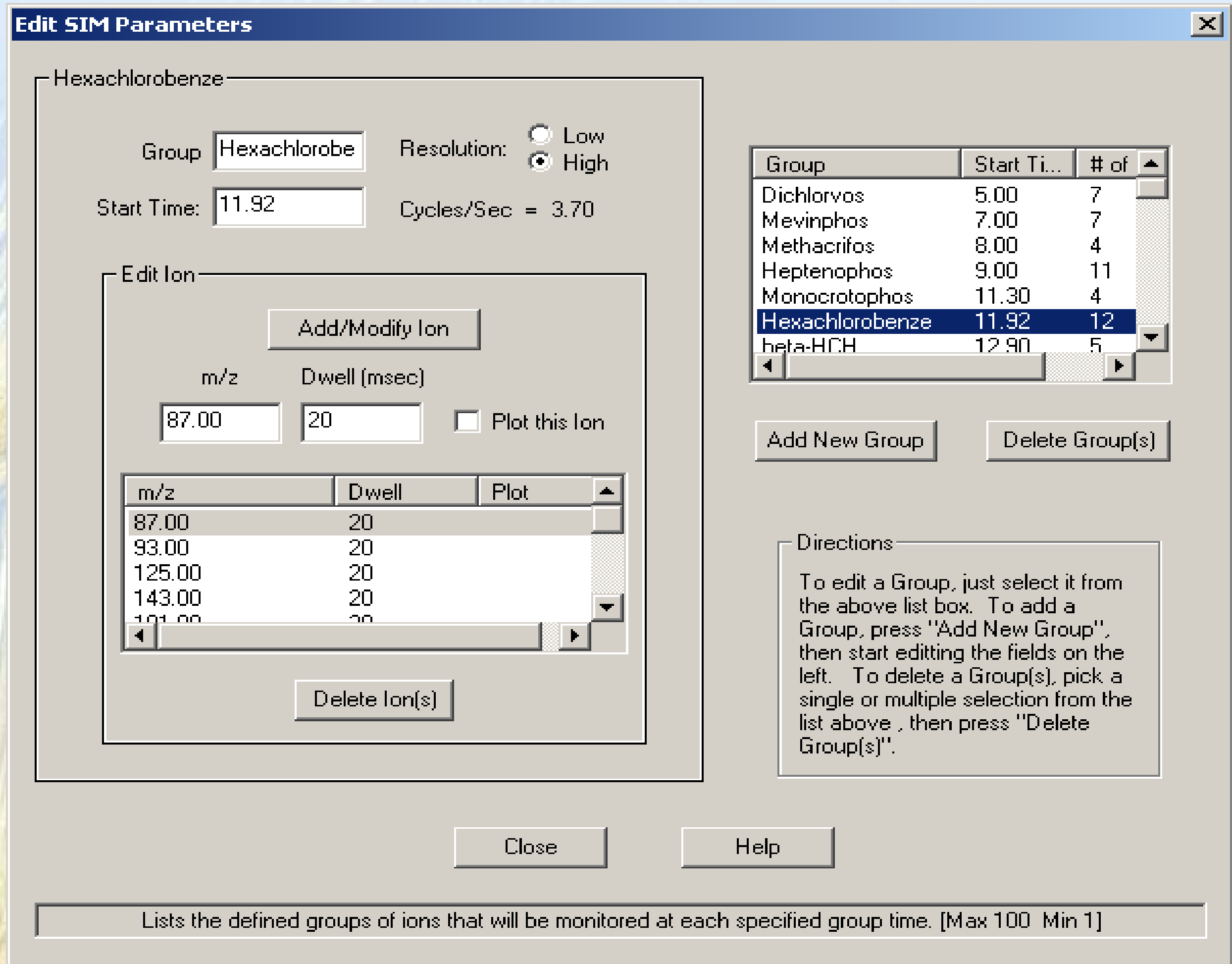
MS/MS

TOF

GC Scope expansion

- SIM methods using single quadrupole MS detectors
- Target ion
- Three qualifiers
- Using retention time locking for consistent retention times
- 200 pesticides in one run

Method



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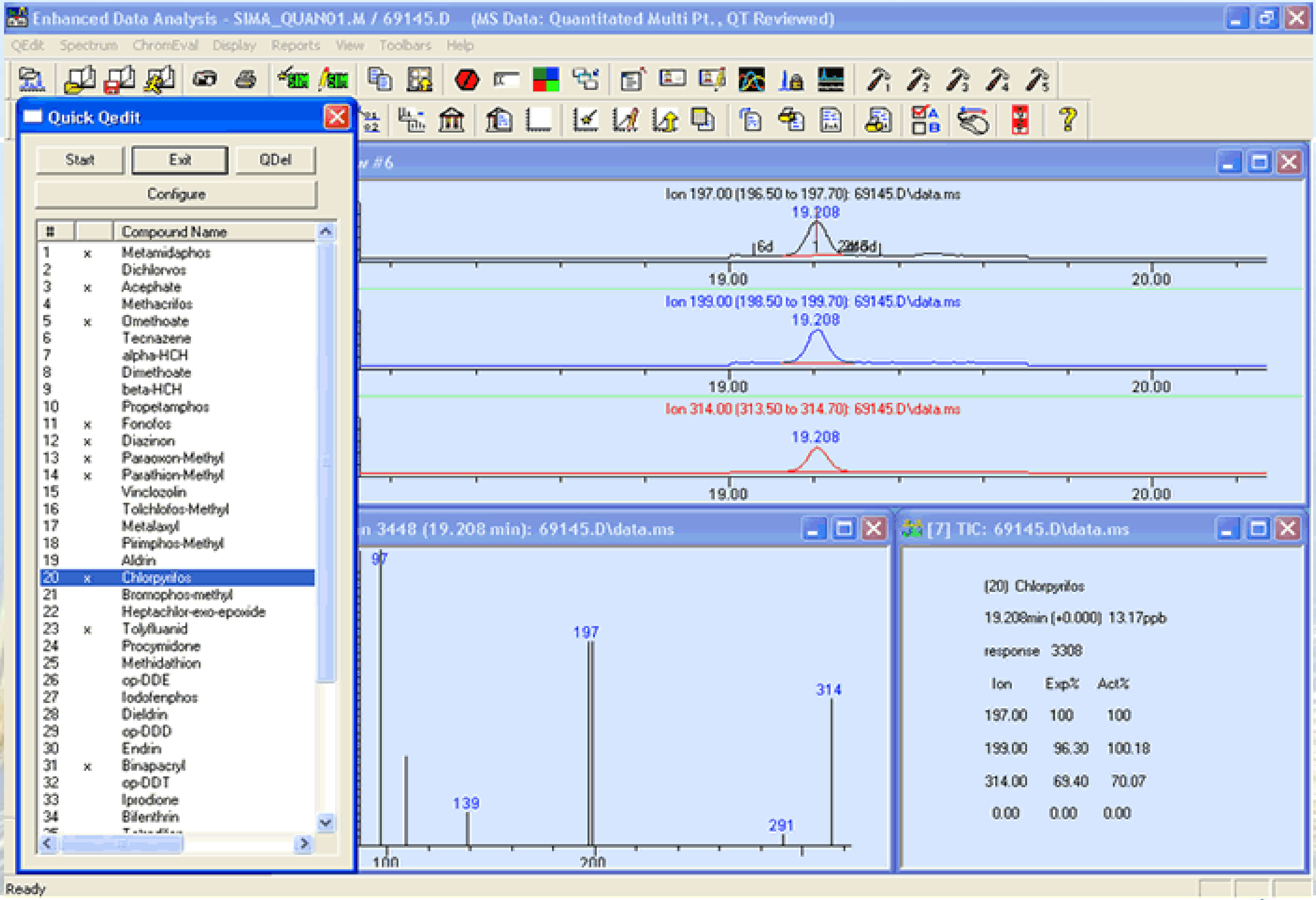

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Recovery work

		Mean	s	% RSD
1	Isoproturon	75.0	20.7	27.6
2	**Nicotine**	**65.7**	**27.7**	**42.1**
3	Propham	102.9	9.2	9.0
4	MCPA Methyl ester	103.4	10.7	10.3
5	Ethoprophos	99.4	6.3	6.4
6	Cyanophos	98.8	7.8	7.9
7	Isazofos	99.2	6.1	6.1
8	Endosulfan ether	94.8	8.2	8.6
9	Cyanazine	84.9	16.8	19.8
10	Chlorthal dimethyl	96.6	6.5	6.7
11	Tetraconazole	109.7	12.2	11.1
12	**Sulfur**	**88.8**	**63.1**	**71.1**
13	Isodrin	88.5	7.9	9.0
14	Endosulfan lactone	91.9	7.1	7.7

Recovery

		Mean	s	% RSD
15	Metazachlor	91.9	9.9	10.8
16	Paclobutrazol	108.0	8.8	8.2
17	Napropamide	100.3	6.1	6.1
18	Picoxystrobin	99.0	9.1	9.2
19	Profenophos	104.4	9.3	8.9
20	Buprofezin	95.2	3.2	3.4
21	Flamprop isopropyl	99.1	6.5	6.5
22	Cyanofenphos	101.3	6.0	6.0
23	Lenacil	100.4	14.6	14.6
24	Trifloxystrobin	104.6	17.1	16.3
25	Epoxyconazole	105.0	17.2	16.4
26	Bifenox	111.2	26.8	24.1
27	Pyridaben	101.8	8.0	7.9
28	Fenbuconazole	97.4	15.0	15.4

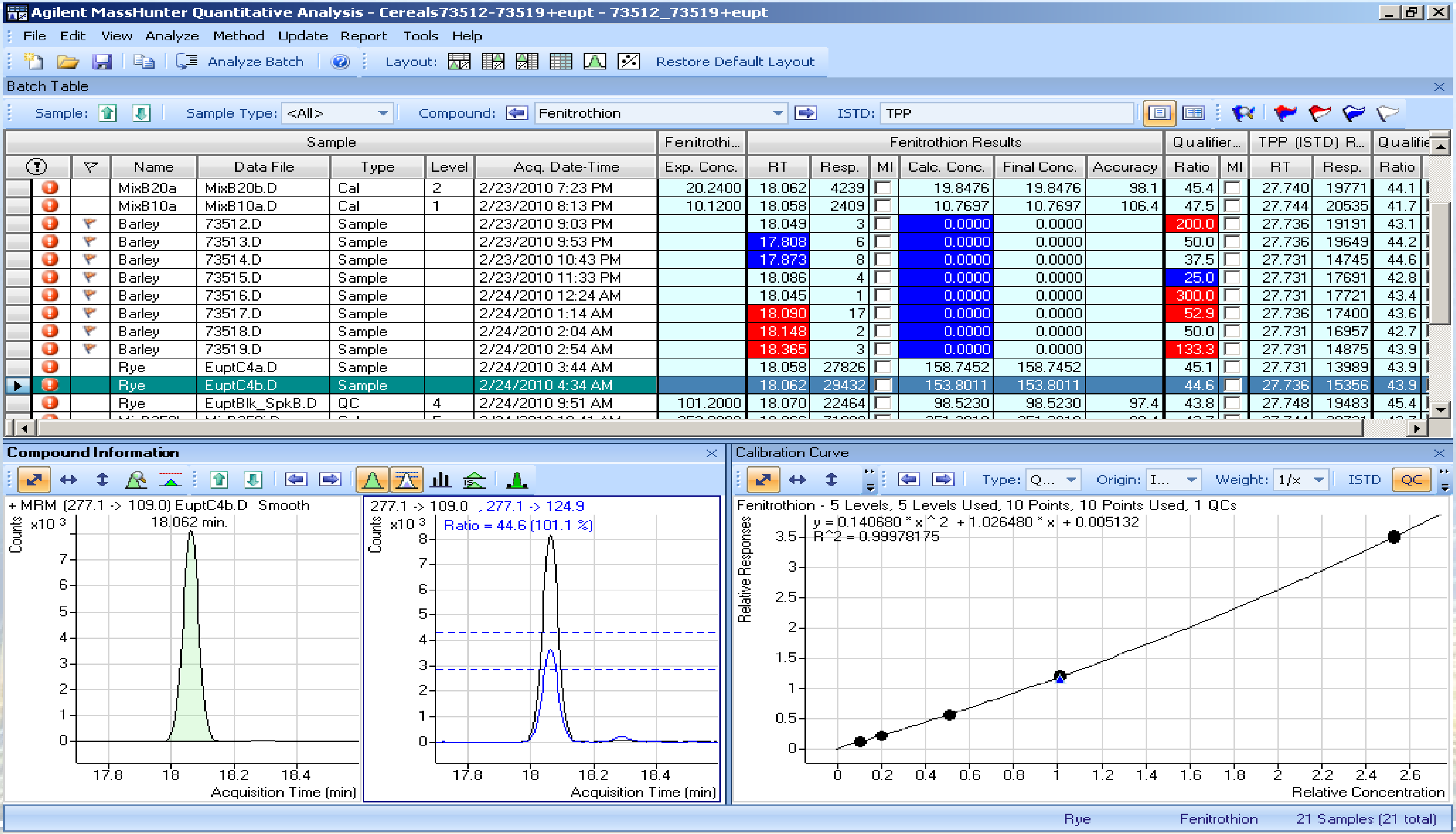
Pro’s and cons

- Quantitation possible for overlapping peaks.
- Getting structural information
- Very good sensitivity – down to 10ppb
- Dependent on molecules fragmenting to give unique ions
- Prone to matrix effects.

Increasing selectivity and sensitivity

- GC-MS/MS
- All the advantage of SIM methods including retention time locking.
- Greater selectivity – One transition for quantitation and one for confirmation
- Increased sensitivity – can now see 1ppb easily in food samples

Data



Name	Data File	Type	Level	Acq. Date-Time	Fenitrothi... Exp. Conc.	Fenitrothion Results RT	Resp.	MI	Calc. Conc.	Final Conc.	Accuracy	Qualifier... Ratio	MI	TPP (ISTD) R... RT	Resp.	Qualifie... Ratio
MixB20a	MixB20b.D	Cal	2	2/23/2010 7:23 PM	20.2400	18.062	4239		19.8476	19.8476	98.1	45.4		27.740	19771	44.1
MixB10a	MixB10a.D	Cal	1	2/23/2010 8:13 PM	10.1200	18.058	2409		10.7697	10.7697	106.4	47.5		27.744	20535	41.7
Barley	73512.D	Sample		2/23/2010 9:03 PM		18.049	3		0.0000	0.0000		200.0		27.736	19191	43.1
Barley	73513.D	Sample		2/23/2010 9:53 PM		17.808	6		0.0000	0.0000		50.0		27.736	19649	44.2
Barley	73514.D	Sample		2/23/2010 10:43 PM		17.873	8		0.0000	0.0000		37.5		27.731	14745	44.6
Barley	73515.D	Sample		2/23/2010 11:33 PM		18.086	4		0.0000	0.0000		25.0		27.731	17691	42.8
Barley	73516.D	Sample		2/24/2010 12:24 AM		18.045	1		0.0000	0.0000		300.0		27.731	17721	43.4
Barley	73517.D	Sample		2/24/2010 1:14 AM		18.090	17		0.0000	0.0000		52.9		27.736	17400	43.6
Barley	73518.D	Sample		2/24/2010 2:04 AM		18.148	2		0.0000	0.0000		50.0		27.731	16957	42.7
Barley	73519.D	Sample		2/24/2010 2:54 AM		18.365	3		0.0000	0.0000		133.3		27.731	14875	43.9
Rye	EuptC4a.D	Sample		2/24/2010 3:44 AM		18.058	27826		158.7452	158.7452		45.1		27.731	13989	43.9
Rye	EuptC4b.D	Sample		2/24/2010 4:34 AM		18.062	29432		153.8011	153.8011		44.6		27.736	15356	43.9
Rye	EuptBlk_SpkB.D	QC	4	2/24/2010 9:51 AM	101.2000	18.070	22464		98.5230	98.5230	97.4	43.8		27.748	19483	45.4

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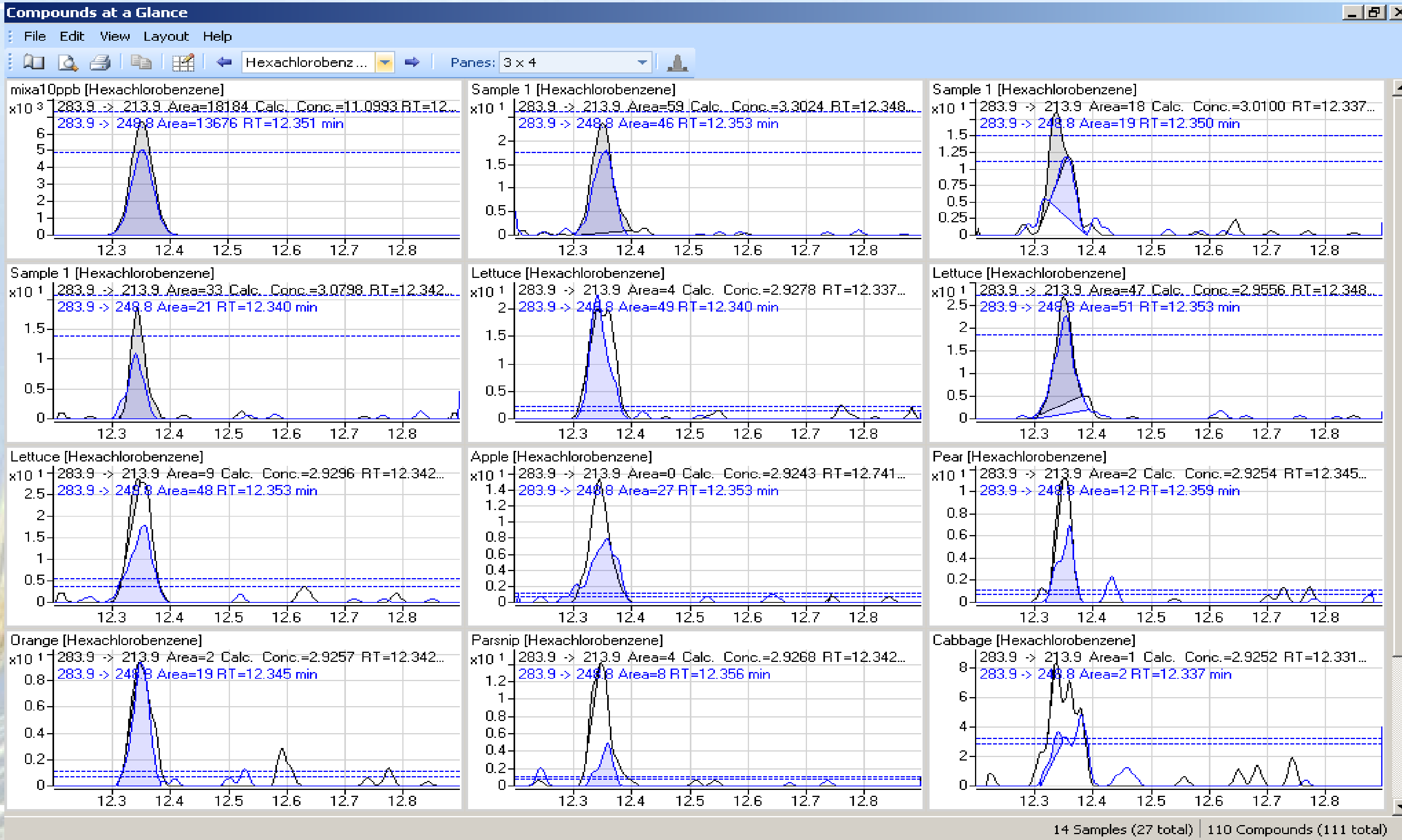
Data


Compounds at a Glance
File Edit View Layout Help
Hexachlorobenz...
Panes: 3 x 4
mixa10ppb [Hexachlorobenzene]
283.9 -> 213.9 Area=18184 Calc. Conc.=11.0993 RT=12...
283.9 -> 248.8 Area=13676 RT=12.351 min
Sample 1 [Hexachlorobenzene]
283.9 -> 213.9 Area=59 Calc. Conc.=3.3024 RT=12.348...
283.9 -> 248.8 Area=46 RT=12.353 min
Sample 1 [Hexachlorobenzene]
283.9 -> 213.9 Area=18 Calc. Conc.=3.0100 RT=12.337...
283.9 -> 248.8 Area=19 RT=12.350 min
Sample 1 [Hexachlorobenzene]
283.9 -> 213.9 Area=33 Calc. Conc.=3.0798 RT=12.342...
283.9 -> 248.8 Area=21 RT=12.340 min
Lettuce [Hexachlorobenzene]
283.9 -> 213.9 Area=4 Calc. Conc.=2.9278 RT=12.337...
283.9 -> 248.8 Area=49 RT=12.340 min
Lettuce [Hexachlorobenzene]
283.9 -> 213.9 Area=47 Calc. Conc.=2.9556 RT=12.348...
283.9 -> 248.8 Area=51 RT=12.353 min
Lettuce [Hexachlorobenzene]
283.9 -> 213.9 Area=9 Calc. Conc.=2.9296 RT=12.342...
283.9 -> 248.8 Area=48 RT=12.353 min
Apple [Hexachlorobenzene]
283.9 -> 213.9 Area=0 Calc. Conc.=2.9243 RT=12.741...
283.9 -> 248.8 Area=27 RT=12.353 min
Pear [Hexachlorobenzene]
283.9 -> 213.9 Area=2 Calc. Conc.=2.9254 RT=12.345...
283.9 -> 248.8 Area=12 RT=12.359 min
Orange [Hexachlorobenzene]
283.9 -> 213.9 Area=2 Calc. Conc.=2.9257 RT=12.342...
283.9 -> 248.8 Area=19 RT=12.345 min
Parsnip [Hexachlorobenzene]
283.9 -> 213.9 Area=4 Calc. Conc.=2.9268 RT=12.342...
283.9 -> 248.8 Area=8 RT=12.356 min
Cabbage [Hexachlorobenzene]
283.9 -> 213.9 Area=1 Calc. Conc.=2.9252 RT=12.331...
283.9 -> 248.8 Area=2 RT=12.337 min
14 Samples (27 total)
110 Compounds (111 total)

Recovery work

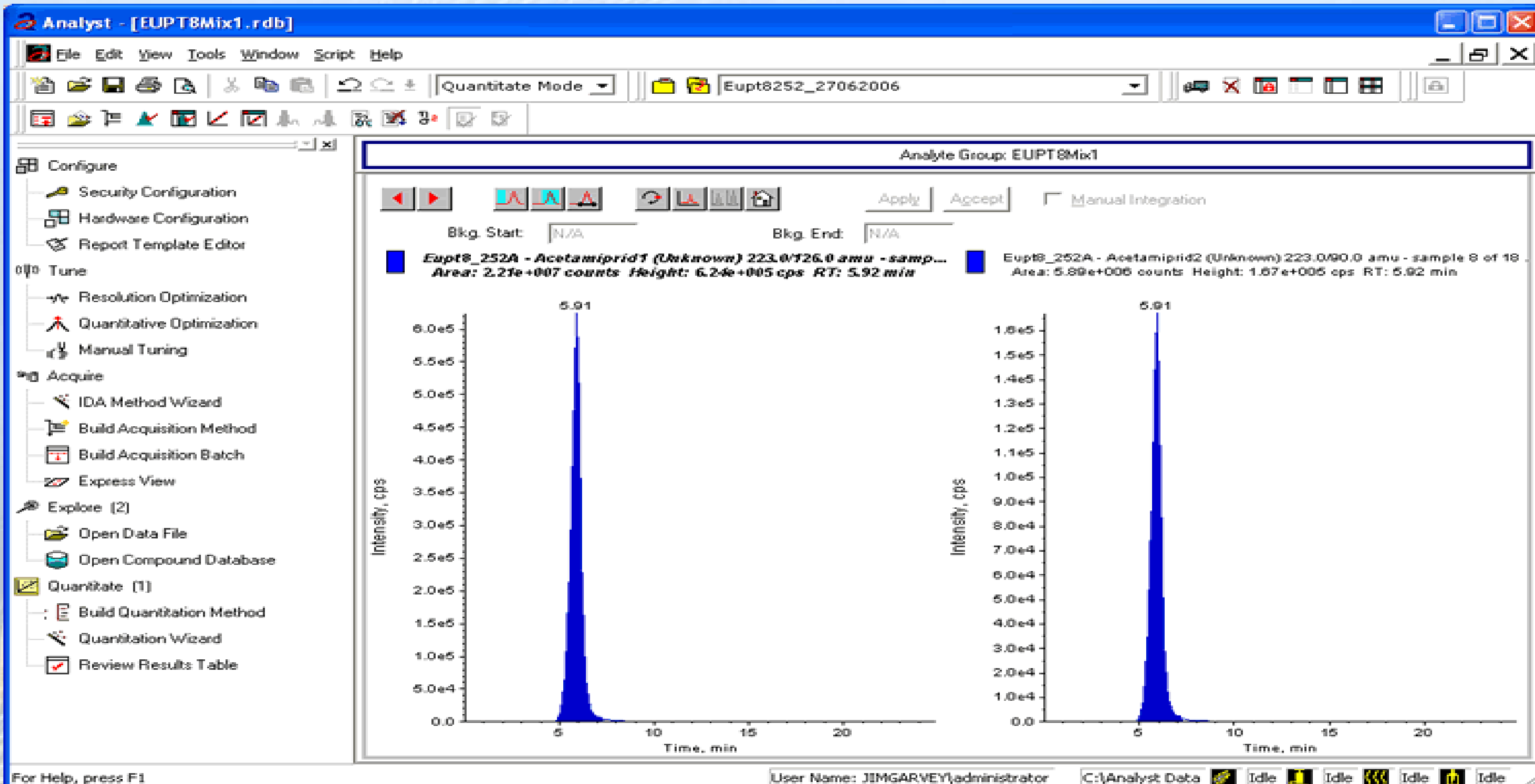
	Mean	s	% RSD
Aldrin	93.8	10.6	11.2
Azinphos ethyl	95.3	10.9	11.4
Azinphos methyl	95.3	12.0	12.6
Bromophos ethyl	96.1	12.1	12.5
Bromophos methyl	96.4	12.7	13.2
Chlorbenzilate	97.1	12.4	12.7
Chlordane cis	96.4	12.3	12.8
Chlordane trans	100.3	13.0	12.9
Chlorfenvinphos	98.0	12.2	12.4
Chlorpyrifos	98.8	10.6	10.7
Chlorpyrifos methyl	96.8	11.1	11.4
Coumaphos	97.0	12.0	12.4
CyfluthrinI	92.0	9.4	10.2

Recovery work

Cypermethrin	96.2	11.0	11.5
Deltamethrin	93.9	19.0	20.3
Diazinon	96.1	12.6	13.1
Dichlorvos	91.0	14.1	15.5
Dieldrin	94.7	15.9	16.8
Dimethoate	104.9	15.2	14.5
Endosulfan alpha	93.5	20.5	21.9
Endosulfan beta	96.8	12.5	12.9
Endrin	97.7	11.4	11.7
Ethion	95.9	11.5	12.0
Fenchlorphos	96.8	12.0	12.4
Fonofos	97.6	12.5	12.8
HCB	88.1	13.4	15.2
Heptachlor	95.5	12.3	12.9
Heptachlor epoxide exo	95.7	12.8	13.4

LC Scope expansion

- MS/MS methods using triple quadrupole MS detectors
- Quantitation transition
- Confirmation transition
- 155 pesticides in one run



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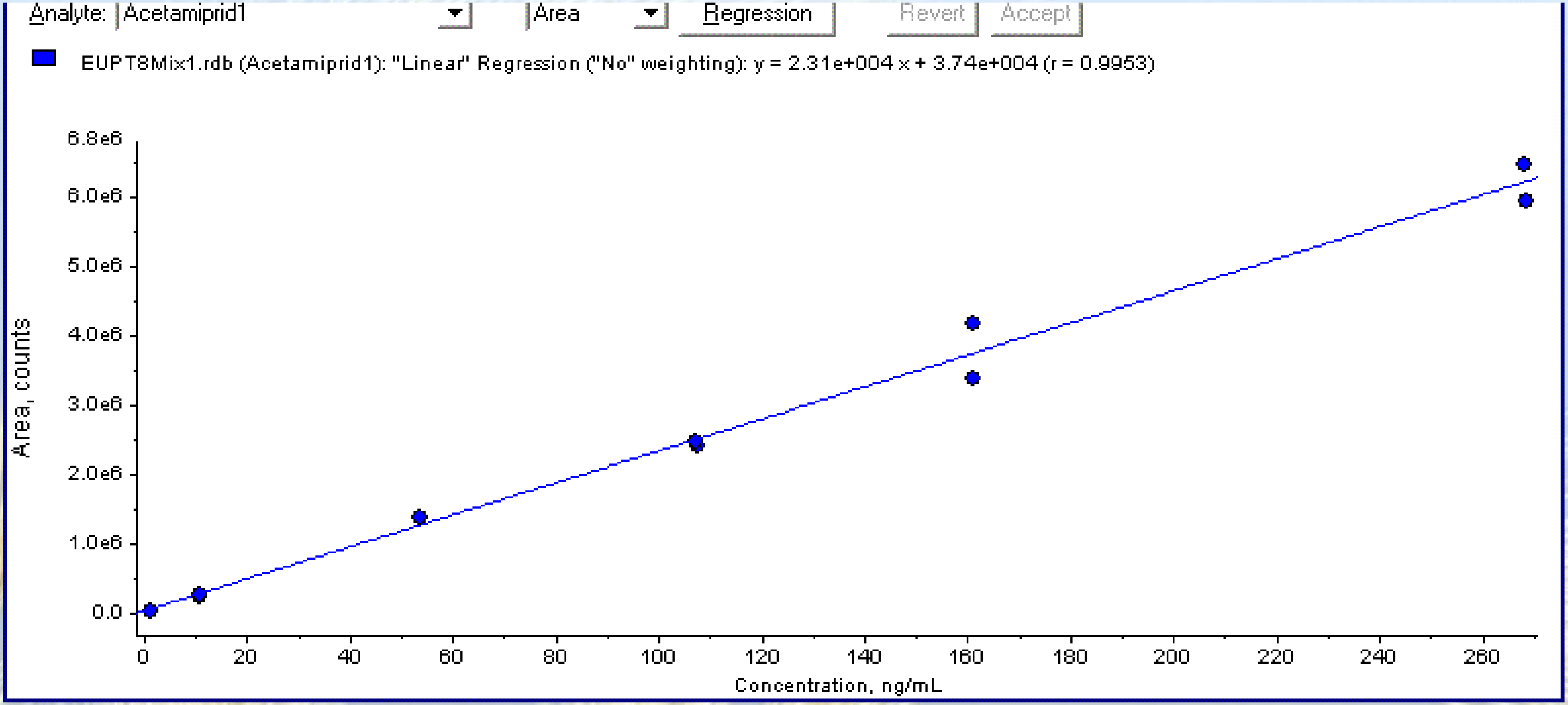
Linearity



Analyte: Acetamiprid1
Area
Regression
Revert
Accept
EUPT8Mix1.rdb (Acetamiprid1): "Linear" Regression ("No" weighting): y = 2.31e+004 x + 3.74e+004 (r = 0.9953)
6.8e6
6.0e6
5.0e6
4.0e6
3.0e6
2.0e6
1.0e6
0.0
Area, counts
0
20
40
60
80
100
120
140
160
180
200
220
240
260
Concentration, ng/mL

Recovery work

Compound	Mean	s	%RSD
Acetamiprid	98.9	10.5	10.6
Aldicarb	113.7	52.3	46.0
Amidosulfuron	114.9	26.0	22.6
Azoxystrobin	99.8	14.4	14.4
Carbaryl	104.9	12.6	12.0
Carbendazim	100.1	10.3	10.2
Carbofuran	103.3	13.2	12.8
Clofentezine	103.6	16.3	15.7
Ethofumesate	104.7	17.9	17.1
Fenhexamid	102.1	11.6	11.4
Fenpropimorph	104.1	11.8	11.4
Fenpyroximate	101.2	16.6	16.4
Flazasulfuron	143.4	47.3	32.9
Hexythiazox	100.1	14.9	14.9
Imazalil	96.3	15.6	16.2

Recovery work

Imidacloprid	103.8	12.8	12.3
Isofenphos	105.7	24.4	23.1
Mepanipyrim	100.3	10.7	10.7
Methamidophos	57.8	12.0	20.8
Methidathion	108.8	26.7	24.6
Methiocarb	103.7	13.6	13.2
Methomyl	99.5	11.7	11.8
Oxamyl	95.6	13.6	14.2
Pymetrozine	73.9	12.6	17.1
Pyridaphenthion	101.6	10.2	10.1
Quinoxyfen	101.3	13.0	12.8
Rotenone	103.7	12.2	11.7
Spinosad	97.7	14.7	15.0
Thiabendazole	100.3	10.3	10.2
Thiacloprid	101.1	13.0	12.8

Improvement

	1999	2010
No of Pesticides	55	350
Analyses / result	3	1
Structural Data	No	Yes
No of standards	13	4

Extraction and clean-up

Method	Extraction	Clean-up
Fruit and veg	(1) Mini-Luke (2) CAN/Buffer	GPC DSPE
Cereals	Ethyl Acetate	GPC
Fats	ACN/Acetone	GPC
Milk	ACN/Buffer	DSPE
Eggs	ACN/Buffer	DSPE
Honey	Mini-Luke	GPC

Older methods

- Used GPC for clean-up
- Long and tedious
- 1.5 days before the samples is ready for analysis

QuEChERS

- Developed by Lehotay et al in 2005
- Two steps
- Extraction with Acetonitrile and buffer
- DSPE clean-up

QuEChERS

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Pro's and cons

- Reduces extraction and clean-up time down to a few minutes
- Applicable to a wide range of pesticides
- ACN as a solvent is not ideal for GC work
- Samples can be quite dirty
- Many variations now exist

Where are we now ?

- MRM's which allow each sample to be analysed for on average 350 – 400 pesticides
- Faster extraction and clean-up
- Recovery for every pesticide in each batch
- Less standard mixes, though more complicated mixes
- Fast GC and LC analysis

Where to now ?

- High mass accuracy MS – TOF
- Excellent screening tool
- Poor for quantitation

What if ?

- We had an instrument which:
- Had a mass accuracy of 1ppm or better
- The quantitation power of a triple quad
- The sensitivity of a triple quad
- Wouldn’t that be good ?

TripleTOF

Potential

- One instrument capable of:
- Pure screening for unknowns
    - And / or
- Targetted analysis
- With
- Quantitation and high sensitivity

A GC equivalent

- Well on the road to development
- Possibly on the market next year
- Initial results are said to be very good

Is there a downside ?

- **Instrumentation is now so sensitive that environmental background levels of pesticides are frequently seen in laboratories.**
- **Methods need to be validated at much lower concentrations which increases the difficulty of the validation**

- **Thank you**

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