Monitoring Pesticides in the Environment: Past, Present and Future – FDA Perspective

National Environmental Monitoring Conference

August 6, 2013

Chris A. Sack, Residue Expert

Food and Drug Administration

Introduction

- Overview
- Regulatory Monitoring
- Program Administration
- Methodology
  - Past
  - Current
  - Future

Overview - Challenges

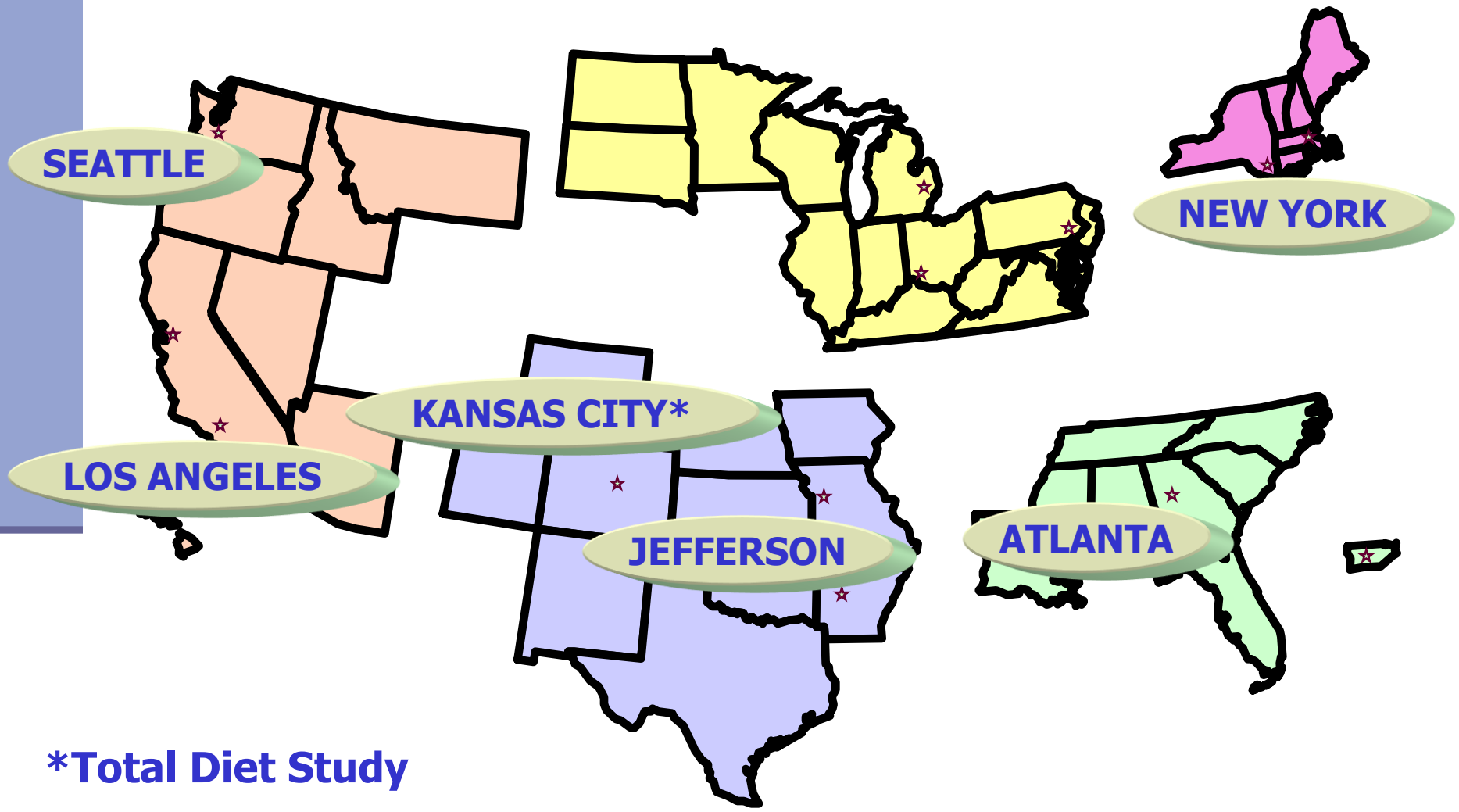
- Food and feed matrices
  - Imports: > 11 million per year
  - Domestic: ???
- Pesticides and other contaminants
  - 1000s that are known
  - Range: 10 ppb - ???
- Analyses
  - Up to 50 samples per day per lab
  - Timeframe: 1 day for imports

Overview - Programs

- Regulatory Monitoring
- Total Diet Study
- Special Assignments

Overview

6 PESTICIDE LABORATORIES

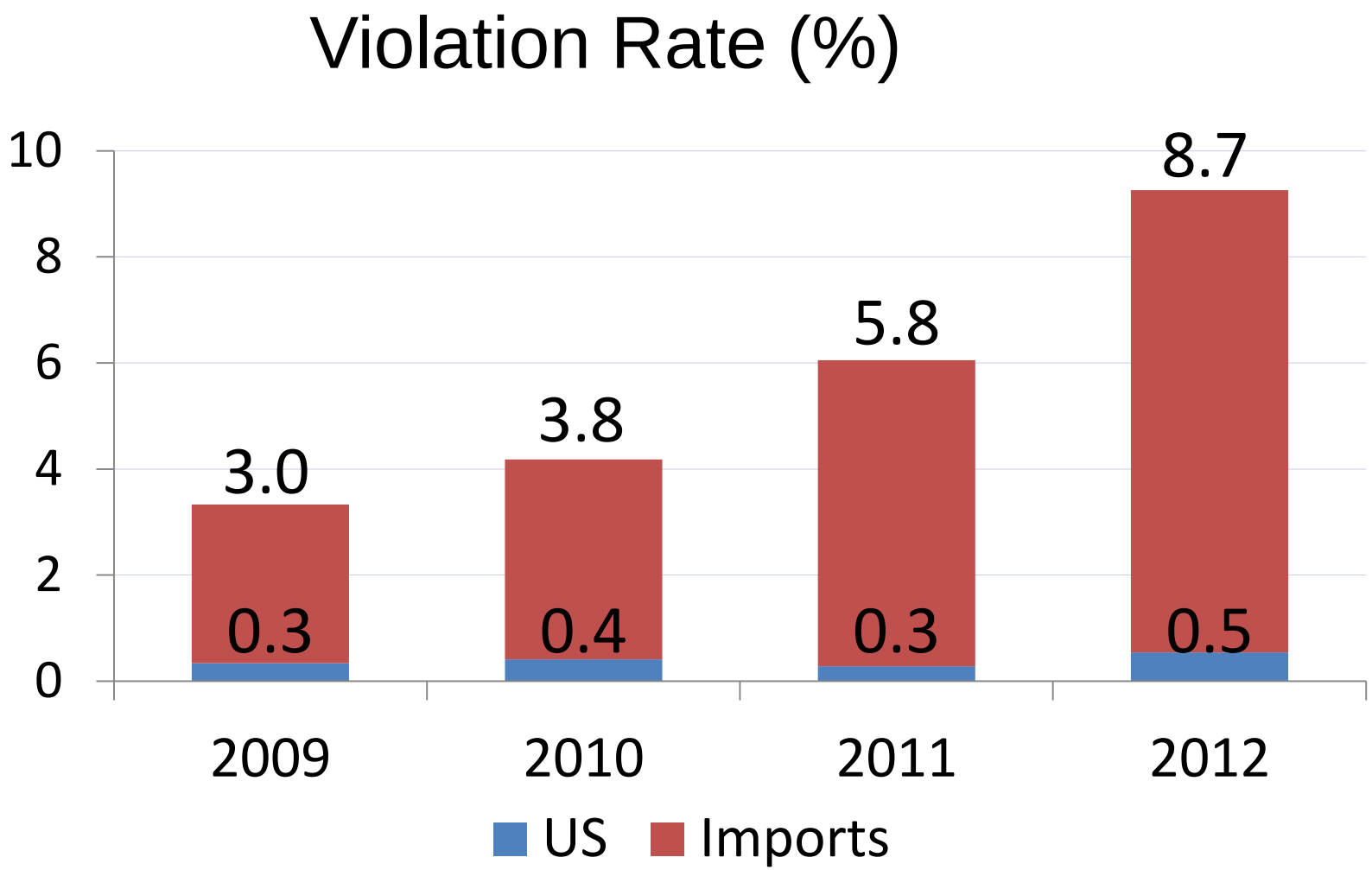


*Total Diet Study

Overview – Regulatory Analysis

- Sample types:
  - Raw agricultural products
  - Processed foods (limited)
  - Spices/botanicals
  - Miscellaneous
- Samples: 5000 - 7000 per year
- Target: ~ 400 pesticides per sample
- Detect: ~ 220 different residues per year

Regulatory Monitoring



Regulatory Monitoring

Year	Violative Residues	Violation Type (%)	
		No MRL	Exceeds MRL
2009	603	96.8	3.2
2010	686	94.5	5.5
2011	996	96.0	4.0
2012	1189	96.8	3.2

Regulatory Monitoring

Most commonly found violative residues (2010–12)

Carbendazim	313	Profenofos	61	DDT	48
Permethrin	179	DCPA	59	Thiophanate-methyl	47
Tricyclazole	104	Endosulfan	57	Pirimiphos methyl	46
Quintozene	100	Pyrimethanil	54	Prochloraz	45
Ethion	70	Procymidone	53	Cypermethrin	41
Chlorpyrifos	70	Lambda-cyhalothrin	51	Tebuconazole	40
Triazophos	69	Buprofezin	51	Carbofuran	40

Regulatory Monitoring

Top violative products (2010–12) *

Product	Vio %	Product	Vio %
Capsicums	41	Spinach	15
Basil	35	Sweet potatoes	15
Cilantro	30	Citrus juice	14
Tea	26	Grapes, raisins	12
Rice products	25	Blackberries	12
Papaya	25	Peppers, hot	11
Taro/dasheen	20	Red beets	11
Prickle pear	20	Leeks	10

* > 50 samples analyzed

Program Administration

- Analyze more samples
  - Increase laboratory efficiency
    - Streamline preparation process
    - Faster methods
    - Modernize data flow - LIMS
  - Uniformity of protocols and methodology
    - National Accreditation to ISO 17025
    - National GC-QQQ, GC-FS, & LC-MS/MS

Program Administration

- Intelligent Sampling
  - Coordination with other federal agencies
    - EPA Pesticide usage data
    - USDA Pesticide Data Program (PDP)
  - Foreign offices in Europe, Asia, Central and South America
  - PREDICT uses FDA historical data to select import samples

Program Administration

- Coordination with states
  - FERN
  - MOUs
- International coordination/outreach
  - Attendance international workshops
  - International Food Safety Training Lab
  - International Capacity Building and Development

Program Administration

Methodology – Past

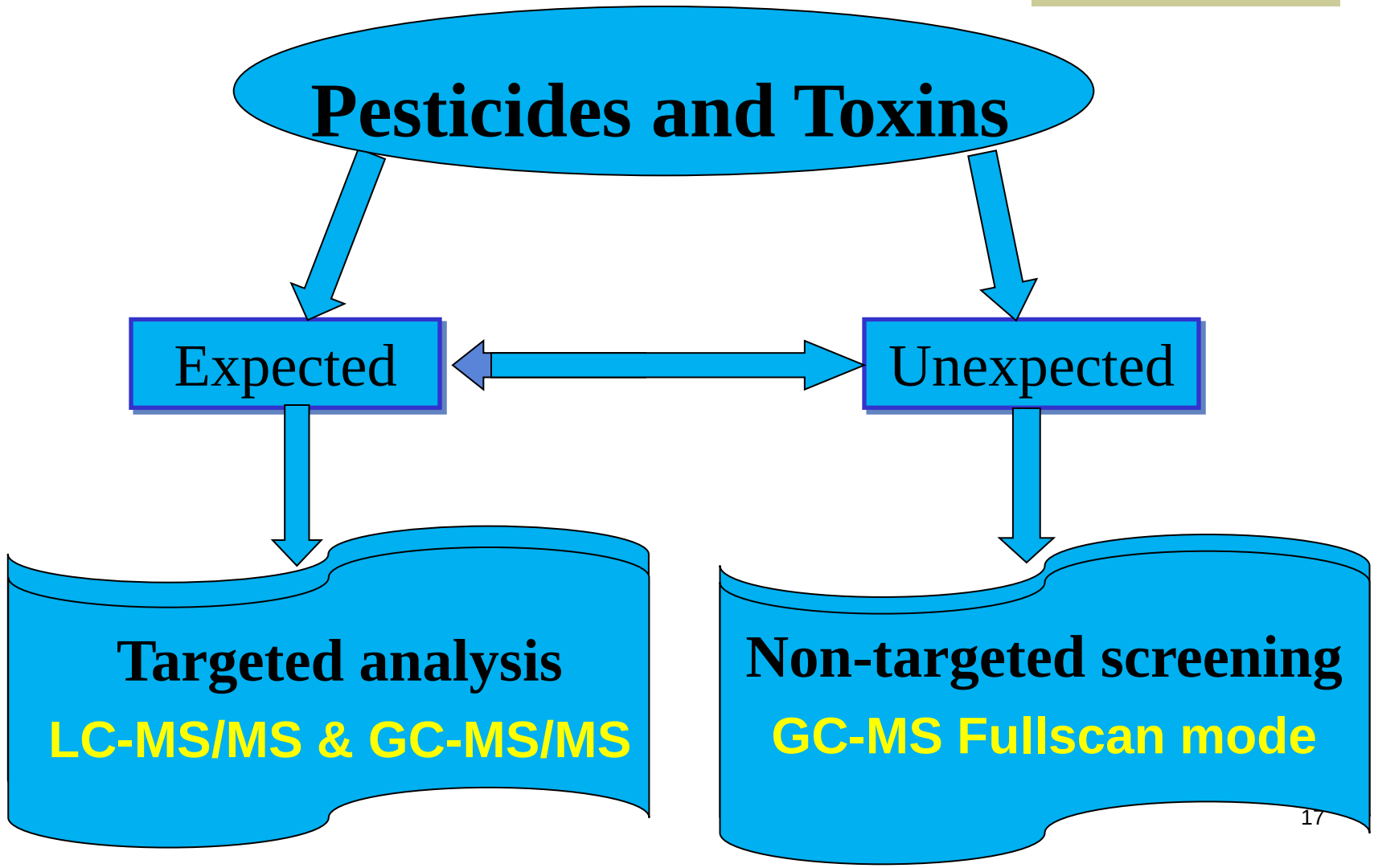
Pesticide Analytical Manual

- Technology of 70s – 80s
- Extraction: Large
- Cleanup: Complex and tedious
- Determination: GC with selective detection
  - Orgranophosphates: FPD
  - Organohalogens: ECD, XSD, ELCD
  - Organonitrogens: NP
- Identification by retention indices

Methodology – Current Extraction/Cleanup

- Acetone extraction (two labs)
  - Sample plus acetone
  - Saltout
  - SPE cleanup
- QuEChERS modified (four labs)
  - Sample plus acetonitrile
  - Saltout
  - SPE cleanup
    - LCMS: PSA only or dilute extract
    - GCMS: PSA + C18 and/or GCB, or equivalent

Methodology – Current Determination



Methodology – Current Determination

- Targeted Quantitative Analysis
  - LC-MS/MS and GC-MS/MS
  - >350 Selected analytes
    - Historical findings
    - Anticipated findings
  - Calibration Standard Mixtures
    - Designed by FDA
    - Prepared by commercial vendor

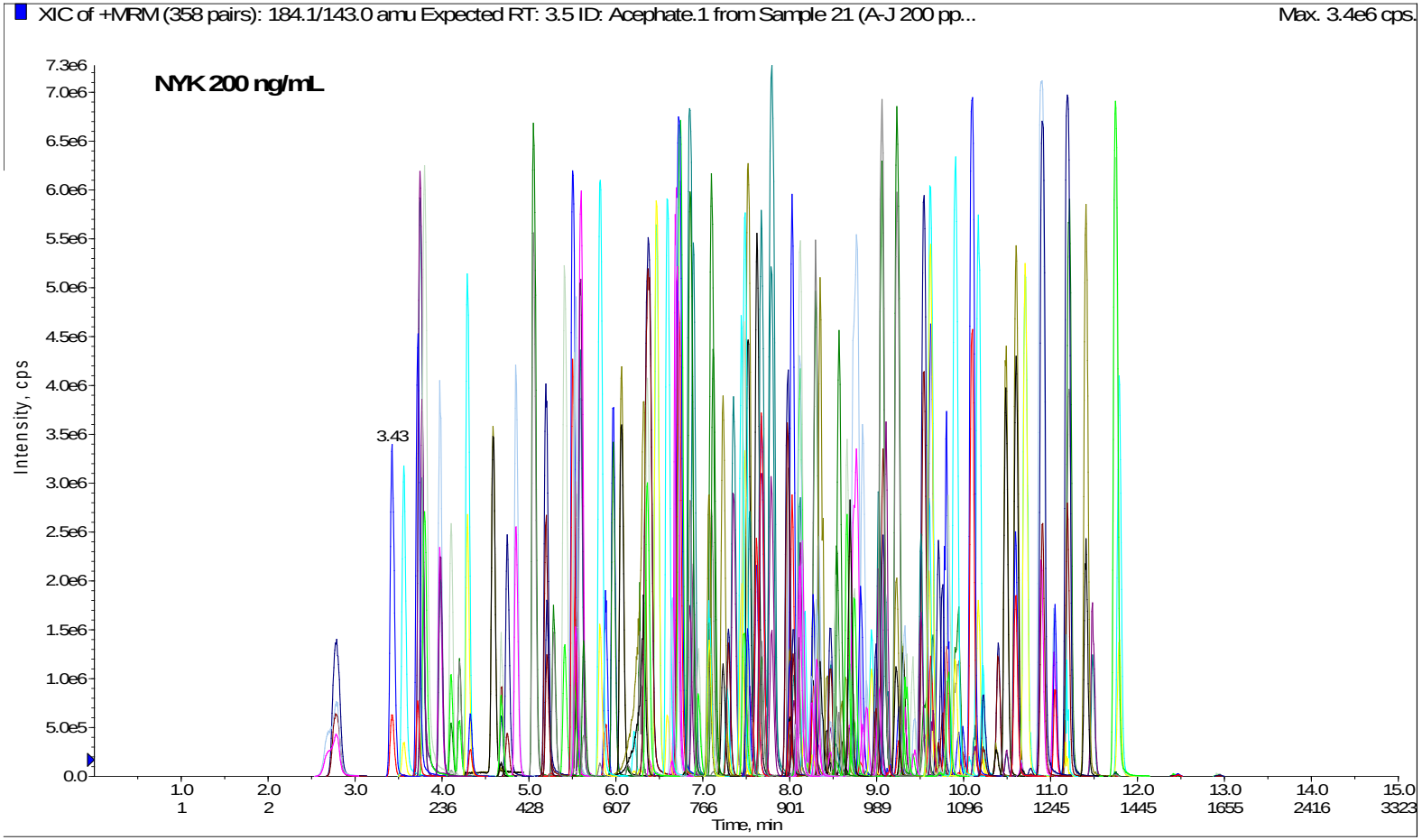
Methodology – Current Determination

- Qualitative Screening Analysis
  - GC-MS in fullscan mode (GC-FS)
  - No standards required
  - Spectral Library Technique

Methodology – Current Determination: LC-MS/MS

- Scope: >240 pesticides in 15 minutes
- Chromatography: reverse phase
- Columns: C_{18}
  - 100 x 2.1 mm, ~3 µm
  - 50 cm x 4.6 mm, 1.9 µm
- Mass Spectrometer parameters
  - Ionization: Positive electrospray
  - Detection: scheduled MRM (two transitions)
- Detection Limit: < 10 ppb for most compounds
- JAFC 2011, Vol 59, pp 6383ff

Methodology – Current Determination: LC-MS/MS



Standard containing 190 pesticides

Methodology – Current Determination: LC-MS/MS

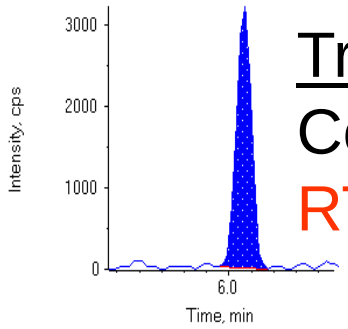
Pesticide	PPB	Pesticide	PPB
Boscalid	128	Novaluron	9
Pyraclostrobin	54	Hexythiazox	5
Cyprodinil	47	Spiromesifen	2
Azoxystrobin	45	Propiconazole	1
Pyrimethanil	41	Acetamiprid	1
Chlorantranilaprole	23	Carbendazim	1
Fludioxinil	22	Methomyl	0.7
Fenhexamid	20	Thiophanate methyl	0.5
Bifenazate	9	Methoxyfenozide	0.3
Myclobutanil	9	Fenpyroximate	0.2

Residues found in composite of 3 strawberry samples

Methodology – Current

Determination: LC-MS/MS

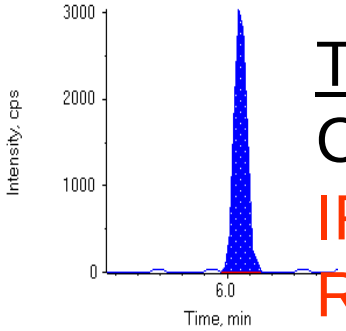
Sample



Trasistion-1

Conc: 0.3 ng/g

RT: 6.07



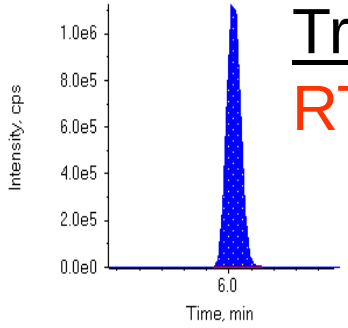
Transition-2

Conc: 0.3 ng/g

IRATIO 85*

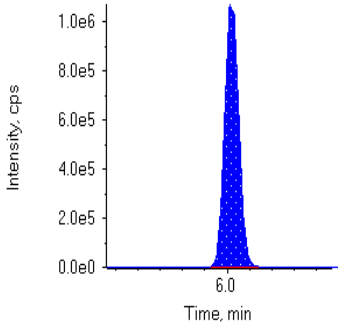
RT 6.06

Standard 50 ng/mL



Transition-1

RT 6.02



Transition-2

IRATIO 99*

RT 6.02

Methoxyfenozide in strawberry @ 0.3 ppb

Methodology – Current Determination: GC-MS/MS

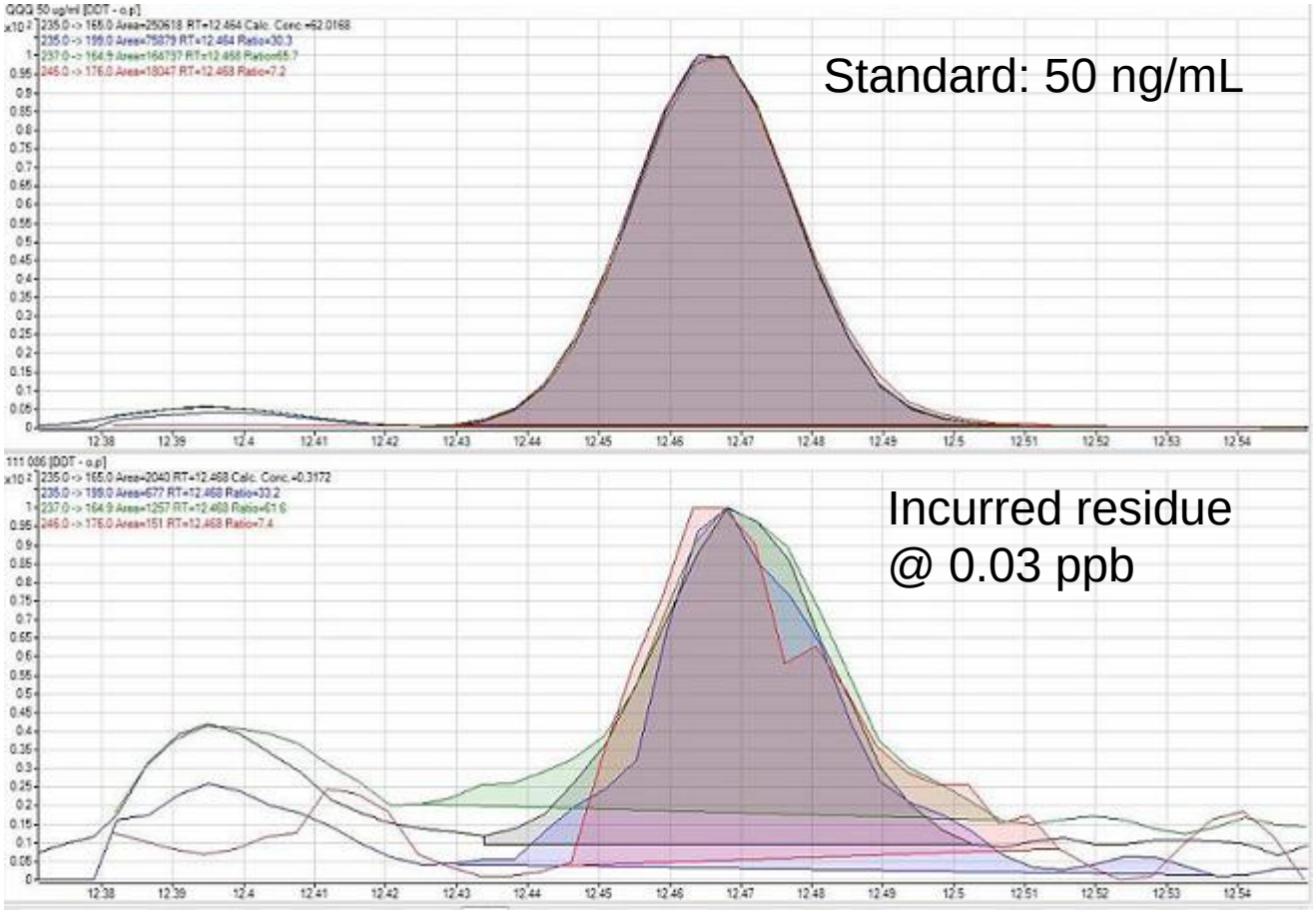
- Scope: > 200 pesticides in 20 minutes
- Agilent 6890GC
  - Retention time locking
  - Backflushing mid column
- Agilent 7000 MS/MS
  - MRM (2-4 transitions/compound)
- Detection Limit: 1 - 10 ppb for most analytes
- USFDA Lab Information Bulletin 4521

Methodology – Current Determination: GC-MS/MS

Pesticide	PPB	Pesticide	PPB
THPI	237	Bifenthrin	7
Boscalid	128	Quinoxyfen	3
Pyraclostrobin	54	Spiromesifen	2
Cyprodinil	47	Propiconazole	1
Pyrimethanil	41	Fenpropathrin	0.7
Azoxystrobin	37	Biphenyl	0.3
Captan	27	Chlorpyrifos	0.1
Fludioxinil	22	Folpet	0.1
Fenhexamid	20	p,p’-DDE	0.1
Bifenazate	9	p,p’-DDT	0.04
Myclobutanil	9	o,p’-DDT	0.03
Malathion	8		

Residues found in composite of 3 strawberry samples

Methodology – Current Determination: GC-MS/MS

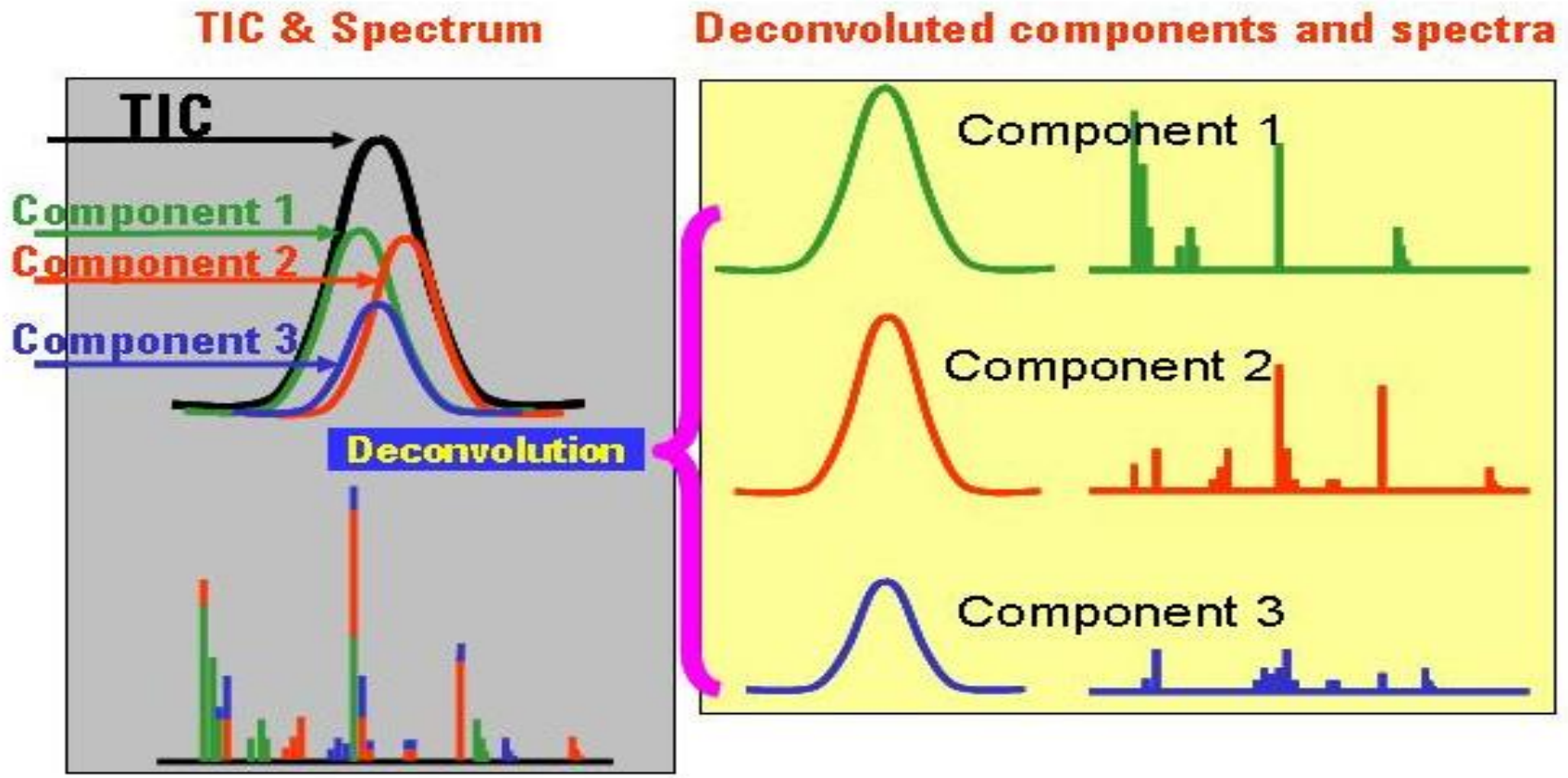


o,p’-DDT in strawberry

Methodology – Current Determination: GC-FS

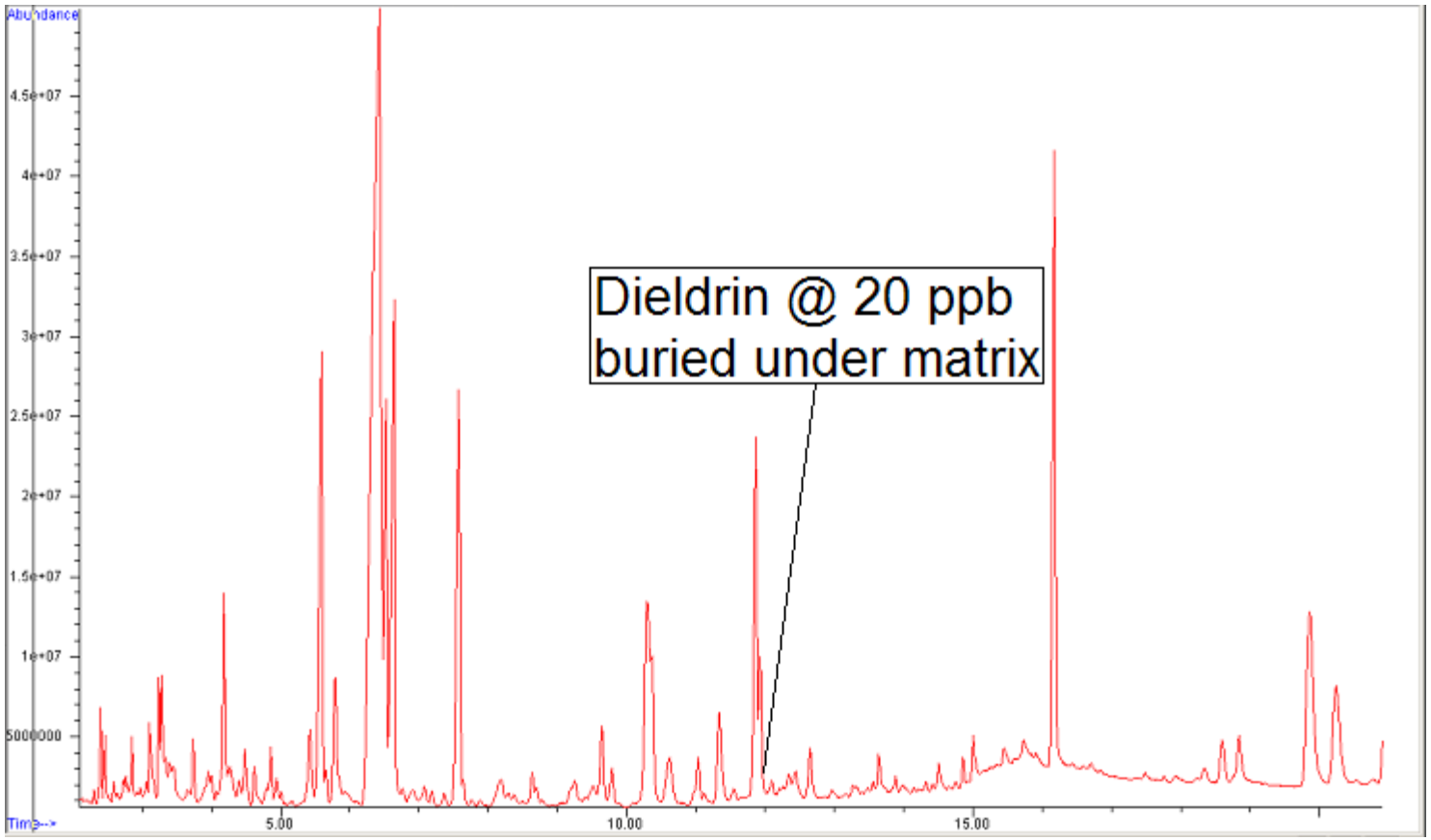
- Scope: > 900 compounds in 20 minutes
- Screen for library matches - no standards
- Agilent GC-MSD pesticide library
- Identification:
  - Spectral matching of AMDIS deconvoluted spectra
  - Retention time

Methodology – Current Determination: GC-FS



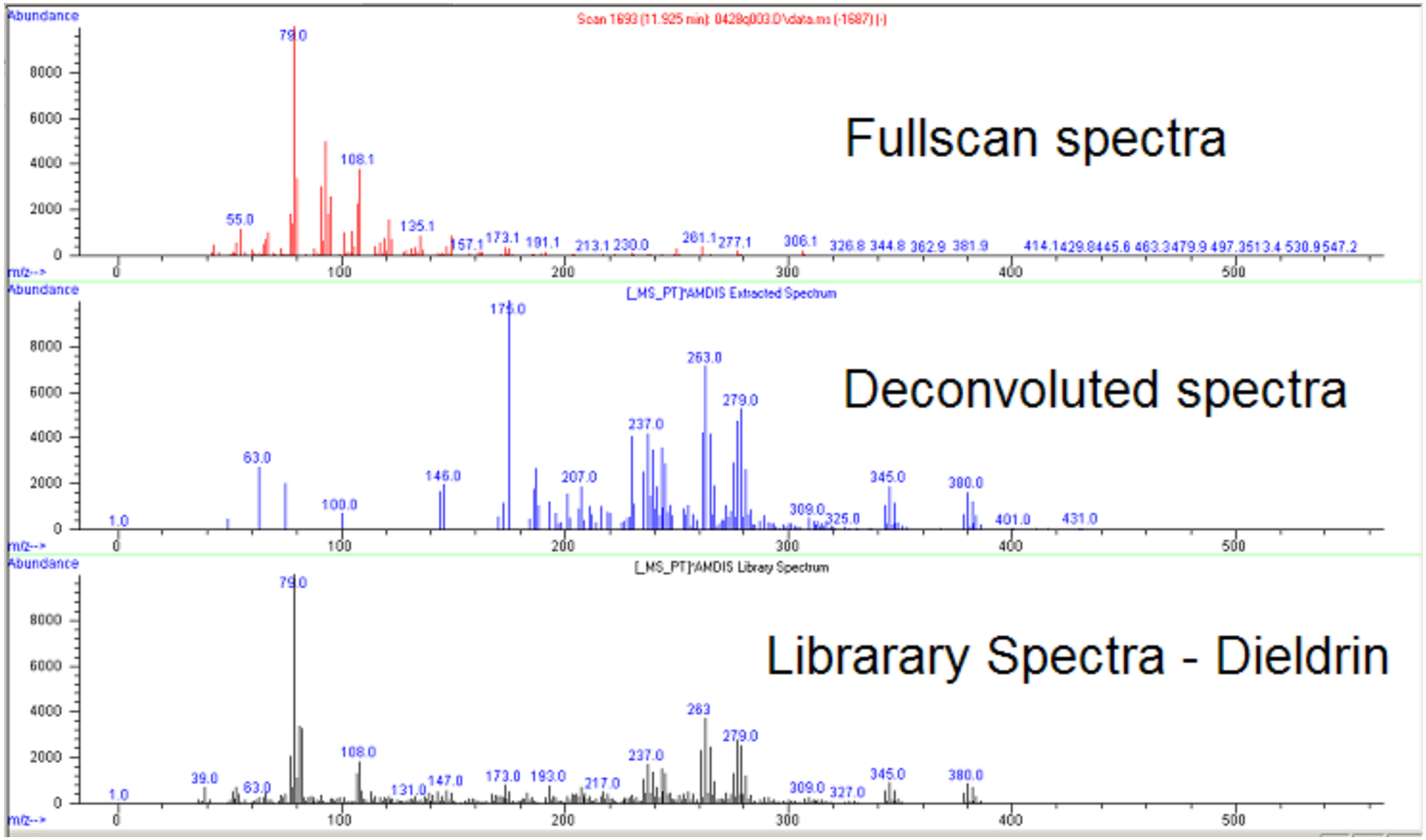
AMDIS Deconvolution

Methodology – Current Determination: GC-FS



Celery fullscan TIC

Methodology – Current Determination: GC-FS



Dieldrin spectra

Methodology – Current Determination: GC-FS

		AMDIS		NIST	
R.T.	Compound Name	Match	R.T. Diff	Reverse Match	Hit Num.
2.5859	Naphthalene-d8	99	-8.0	91	1
2.842	Carvone	75	-8.3	81	2
3.341	EPTC	94	-7.2	89	2
6.977	Anthracene	80	-5.4	88	5
7.9576	Diisobutyl phthalate	75	1.2	84	11
9.214	Di-n-butylphthalate	65	0.8	90	18
9.658	Parathion	82	2.5	76	2
11.925	**Dieldrin**	**79**	**1.5**	**61**	**2**
14.3189	Chrysene-d12	79	-12.1	91	1
14.8515	Bis(2-ethylhexyl)phthalate	96	3.3	85	7
15.7388	Permethrin I	60	6.5	63	2
15.833	Permethrin II	63	7.0	79	
16.518	Cypermethrin II	43	11.7	57	1

Celery deconvolution report

Methodology – Current Determination: GC-FS

Residues discovered and transferred to the LC-MS/MS and GC-MS/MS targeted analyses:

- Fluopicolide
- Spiromesifen
- Spirodiclofen
- Flonicamide
- Chlorfenapyr
- Etoxazole
- Bifenazate
- Etoxazole
- Fenamidone
- Famoxadone
- Quinoxyfen
- Sudan I*

**Food color prohibited in US - not added to target analysis*

Methodology – Future

Residue Screening

- Screening Approach
  - "Semi"- targeted
    - Collect full spectral/RT data
    - Knowns: Libraries
    - Unknowns: Investigate
  - Reduce real-time processing/maintenance
- Technology - HRMS
  - DART
  - LCMS

Methodology – Future

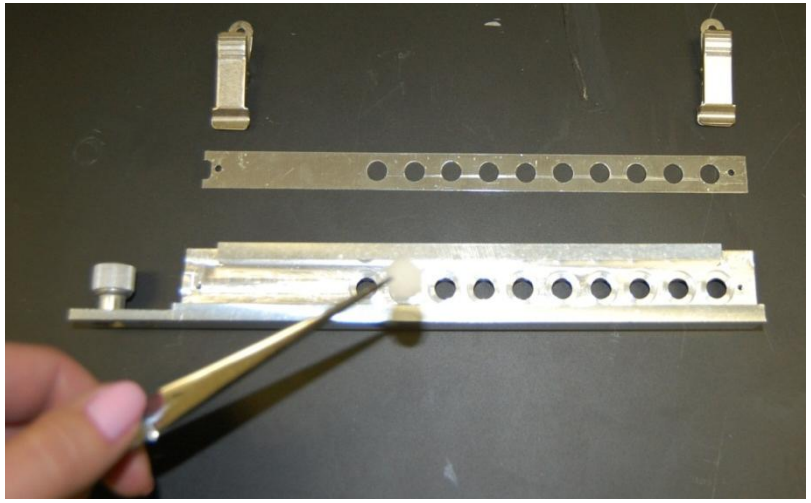
Residue Screening: DART

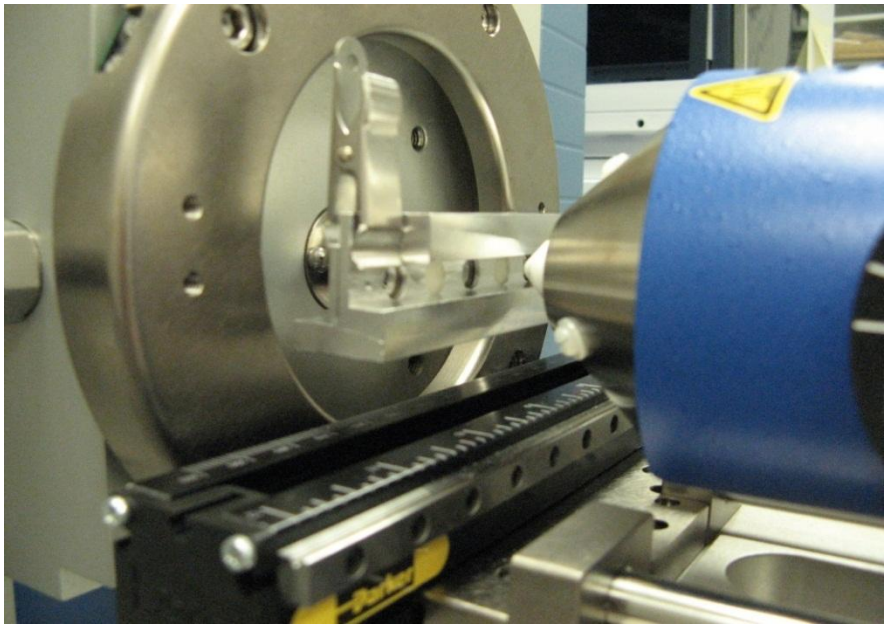
- Method
  - Swab sample
  - Analyze swab by DART-HRMS
- Scope: > 500 pesticides and toxins
- Analysis time: 7.5 minutes/sample
- Implementation:
  - Fast screen of imports at port of entry
  - Forward suspect violations for quantitative analysis

Methodology – Future

Residue Screening: DART

Custom foam rail autosampler

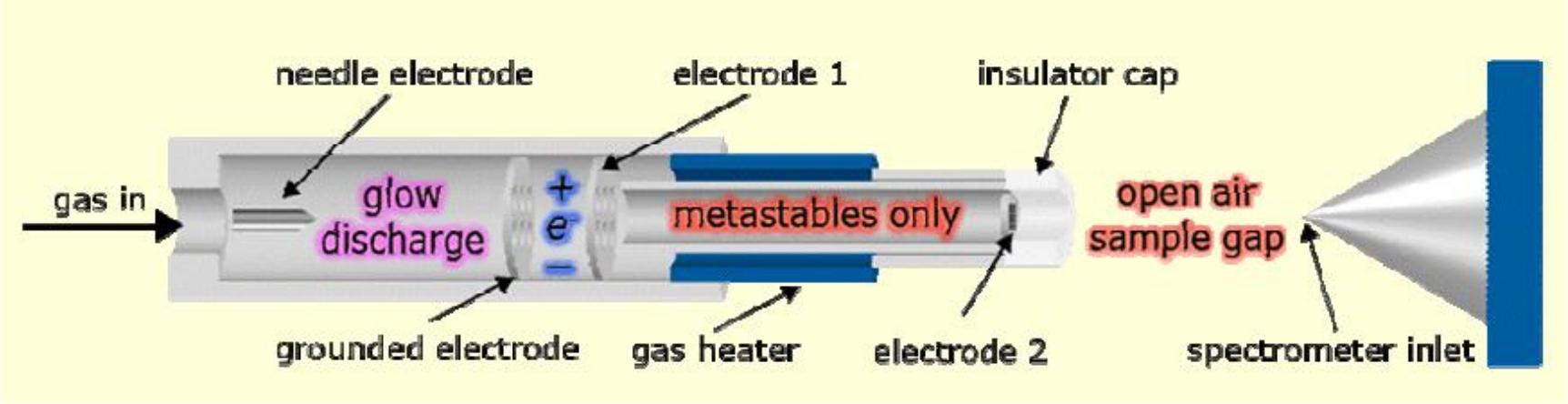




Methodology – Future

Residue Screening: DART

Ionization



$$He(2^3S) + nH_2O \rightarrow [(H_2O)_{n-1} + H]^+ + OH^{\cdot -} + He(1^1S)$$

$$[(H_2O)_n + H]^+ + M \rightarrow [M + H]^+ + nH_2O$$

Methodology – Future

Residue Screening: DART

Thermo Exactive Orbitrap

- Resolution
  - 100,000 @ 1 scan per second
  - 10,000 at 10 scans per second
- Mass accuracy: <1 ppm
- Scan speed: → 10 per second
- Scan range: 50 – 4000 m/z
- Detection Limit: < 10 ng/ml (100 ppb) most compounds

Methodology – Future

Residue Screening: LCMS

- Thermo Exactive Q-Orbitrap
  - LC: Reverse Phase uHPLC
  - Ionization ESI^+
  - MS Library: HR molecular ion & fragments
  - Screen > 600 pesticides @ ≤ 10 ppb
  - Expand to other chemical contaminants: mycotoxins, plant toxins, veterinary drugs, dyes, emerging organic pollutants, and unknown contaminants
- LC-QTOF: under investigation

Conclude

- Continuous Improvement
  - Domestic & international cooperation
  - Smarter sampling
  - Faster sample preparation
  - Screening technologies

Acknowledgements

- David Eide & Michael Smoker (FDA-Kansas City)
- Jon Wong (FDA-CFSAN)
- Greg Mercer & Barb Neuhaus (FDA-Seattle)
- Lorrie Lin & Sara Edison (FDA Forensic Chemistry Center)