



Screening and Quantitation of 215 Pesticides in Honey by an Integrated On-Line Extraction UHPLC-MS/MS System

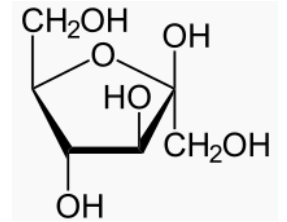
Zicheng Yang and Louis Maljers
September 23, 2015



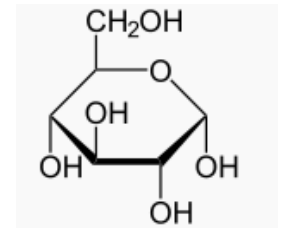
Honey

Nutritional value per 100 g (3.5 oz)

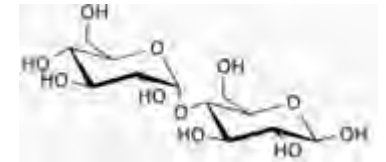
Energy	1,272 kJ (304 kcal)	
Carbohydrates	82.4 g	
Sugars	82.12 g	
Dietary fiber	0.2 g	
Fat	0 g	
Protein	0.3 g	
Vitamins		
Riboflavin (B ₂)	0.038 mg	(3%)
Niacin (B ₃)	0.121 mg	(1%)
Pantothenic acid (B ₅)	0.068 mg	(1%)
Vitamin B ₆	0.024 mg	(2%)
Folate (B ₉)	2 µg	(1%)
Vitamin C	0.5 mg	(1%)
Trace minerals		
Calcium	6 mg	(1%)
Iron	0.42 mg	(3%)
Magnesium	2 mg	(1%)
Phosphorus	4 mg	(1%)
Potassium	52 mg	(1%)
Sodium	4 mg	(0%)
Zinc	0.22 mg	(2%)
Other constituents		
Water	17.10 g	



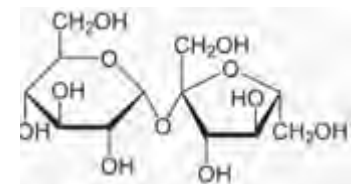
Fructose: 38.2%



Glucose: 31.3%

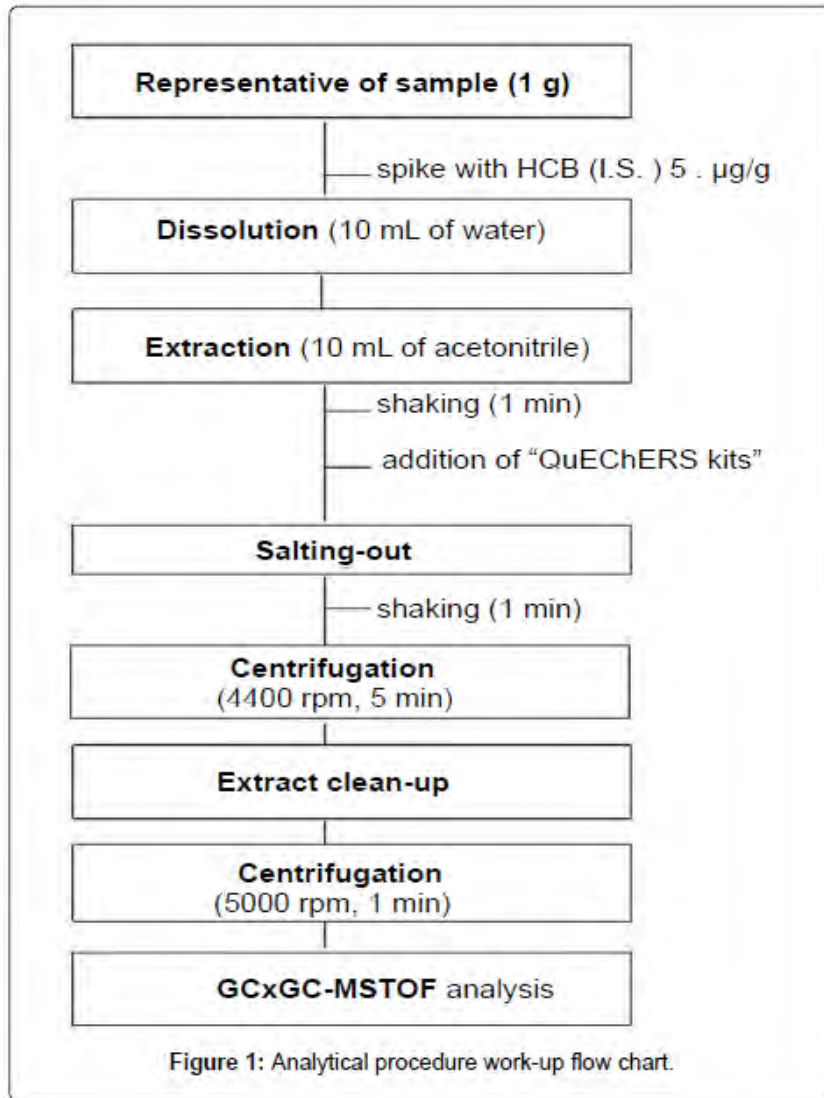


Maltose: 7.1%



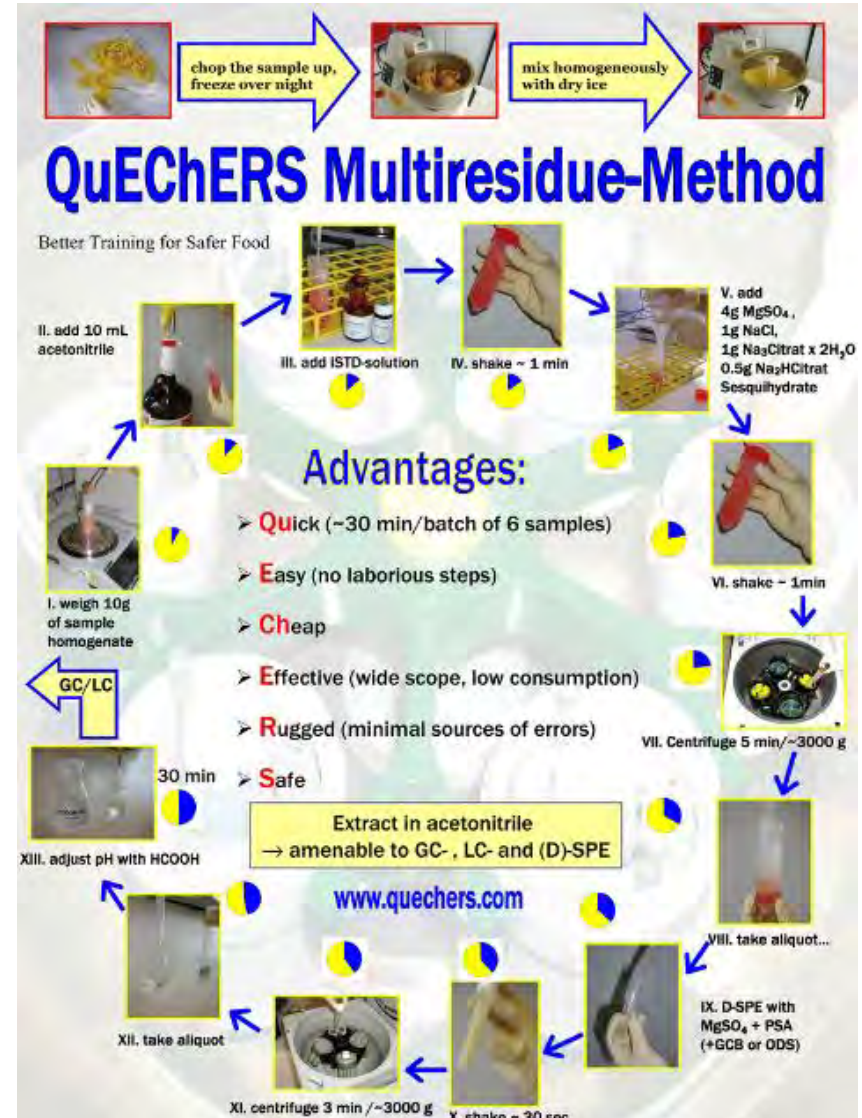
Sucrose: 1.3%

Sample Prep Using QuEChERS



Determination of Pesticide Residues in Honey using the GCxGC-TOFMS Technique

Bargańska et al., J Bioproc Biotech 2014, 4:7
<http://dx.doi.org/10.4172/2155-9821.1000182>

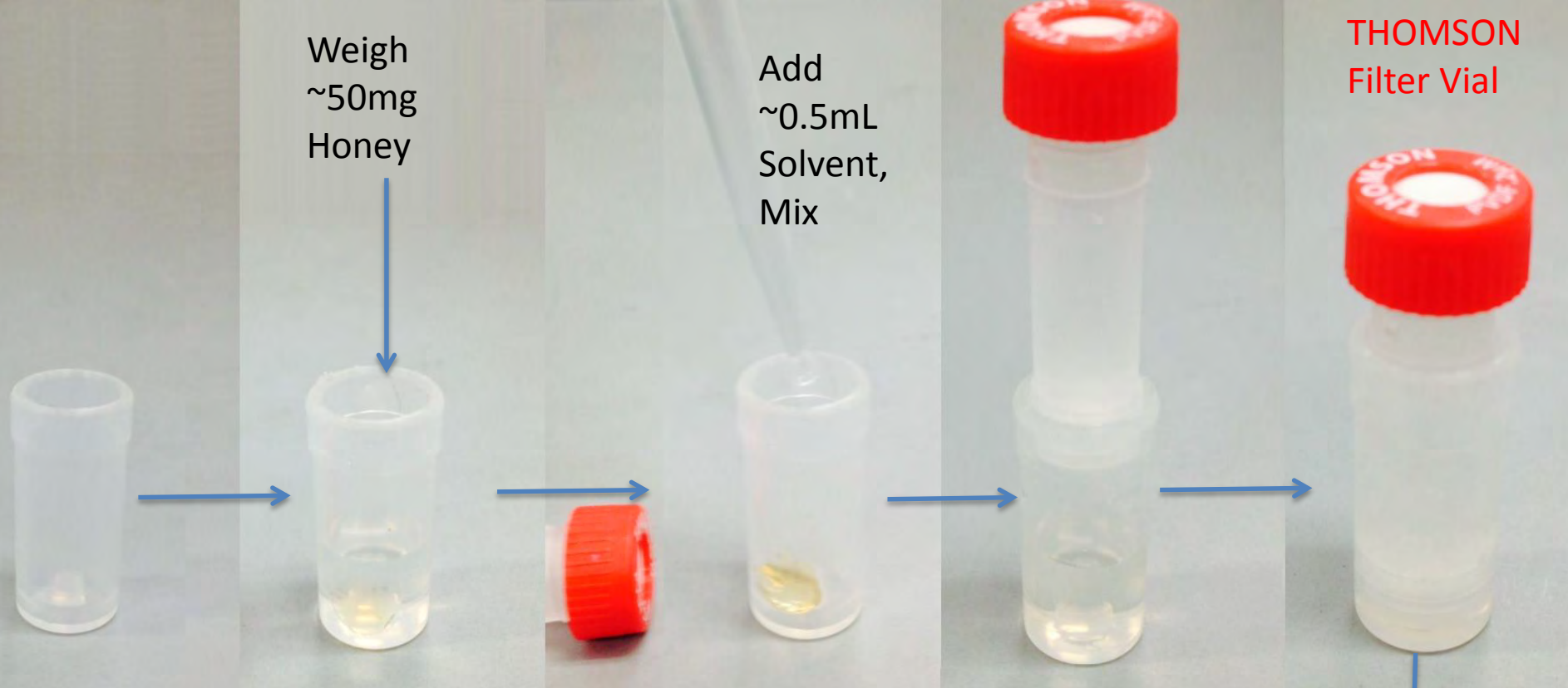


Sample Prep Using SPE

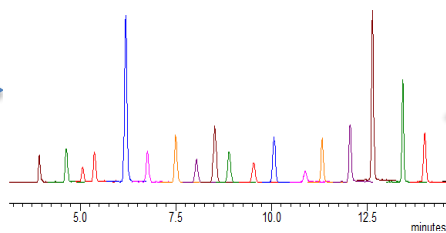


A typical solid phase extraction manifold. The cartridges drip into the chamber below, where tubes collect the effluent. A vacuum port with gauge is used to control the vacuum applied to the chamber.

Condition SEP with MeOH	
Equilibration with water then extracting buffer	
Load sample solution	
Wash with water then Organic/water	
Dry cartridge by vacuum	
Elute with MeOH	
Evaporate to Dryness	
Reconstitute for analysis	

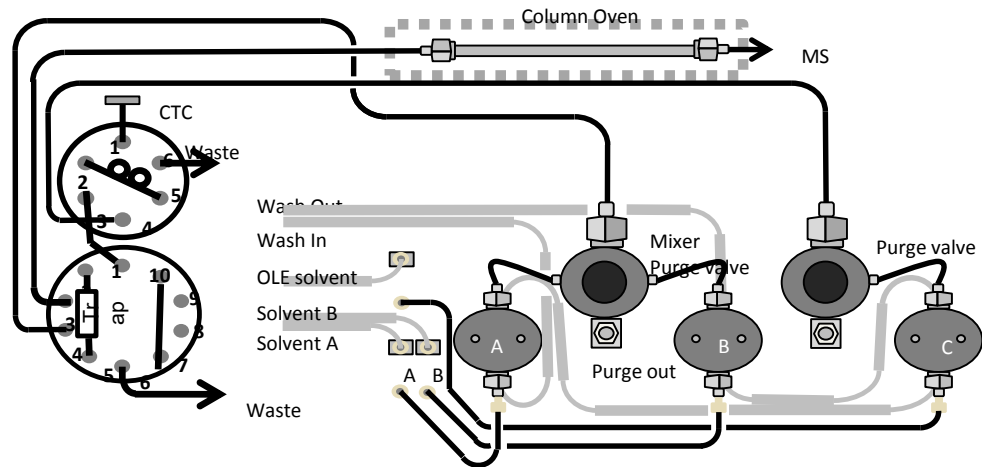


Dilute-Filter-Shoot

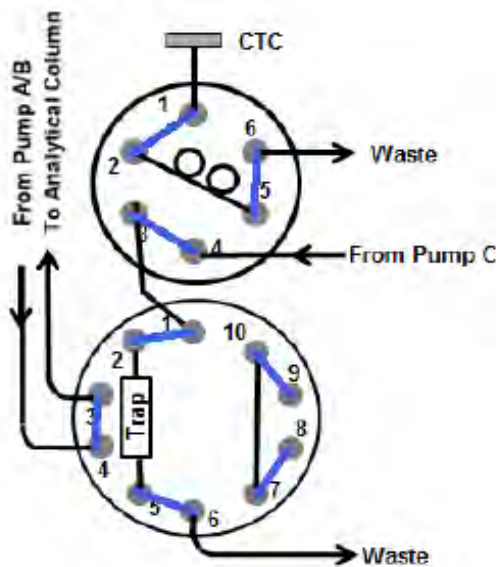


Retention Time (min)	Area	Height	Width	Signal
5.1	10000	1000	0.5	10000
5.5	20000	2000	0.5	20000
6.0	50000	5000	0.5	50000
6.5	100000	10000	0.5	100000
7.0	200000	20000	0.5	200000
7.5	500000	50000	0.5	500000
8.0	1000000	100000	0.5	1000000
8.5	2000000	200000	0.5	2000000
9.0	5000000	500000	0.5	5000000
9.5	10000000	1000000	0.5	10000000
10.0	20000000	2000000	0.5	20000000
10.5	50000000	5000000	0.5	50000000
11.0	100000000	10000000	0.5	100000000
11.5	200000000	20000000	0.5	200000000
12.0	500000000	50000000	0.5	500000000
12.5	1000000000	100000000	0.5	1000000000
13.0	2000000000	200000000	0.5	2000000000
13.5	5000000000	500000000	0.5	5000000000
14.0	10000000000	1000000000	0.5	10000000000
14.5	20000000000	2000000000	0.5	20000000000
15.0	50000000000	5000000000	0.5	50000000000

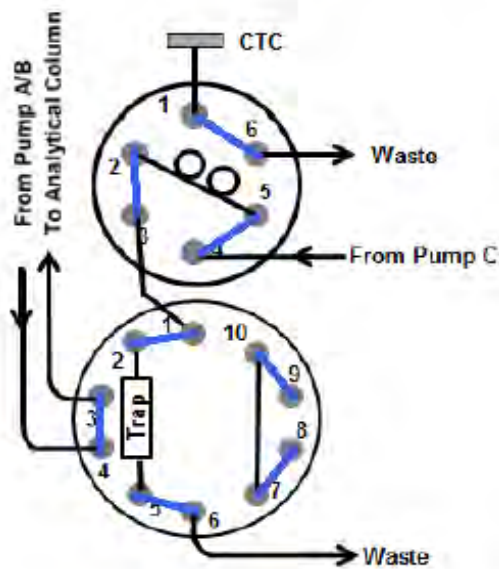
Valves Configuration



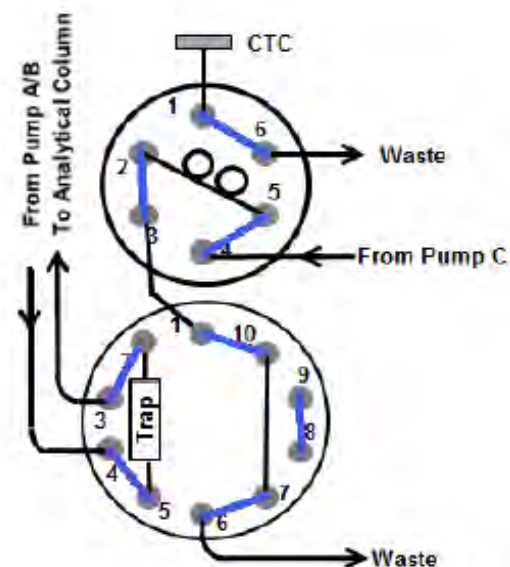
Equilibration Loading



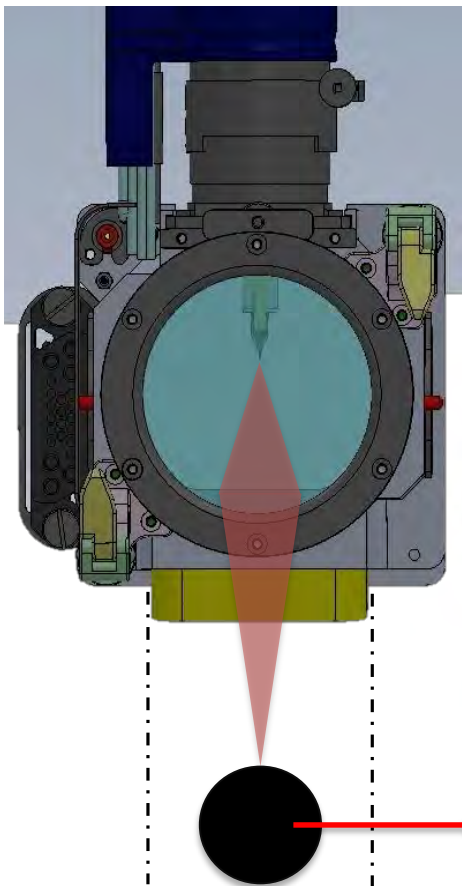
Trapping



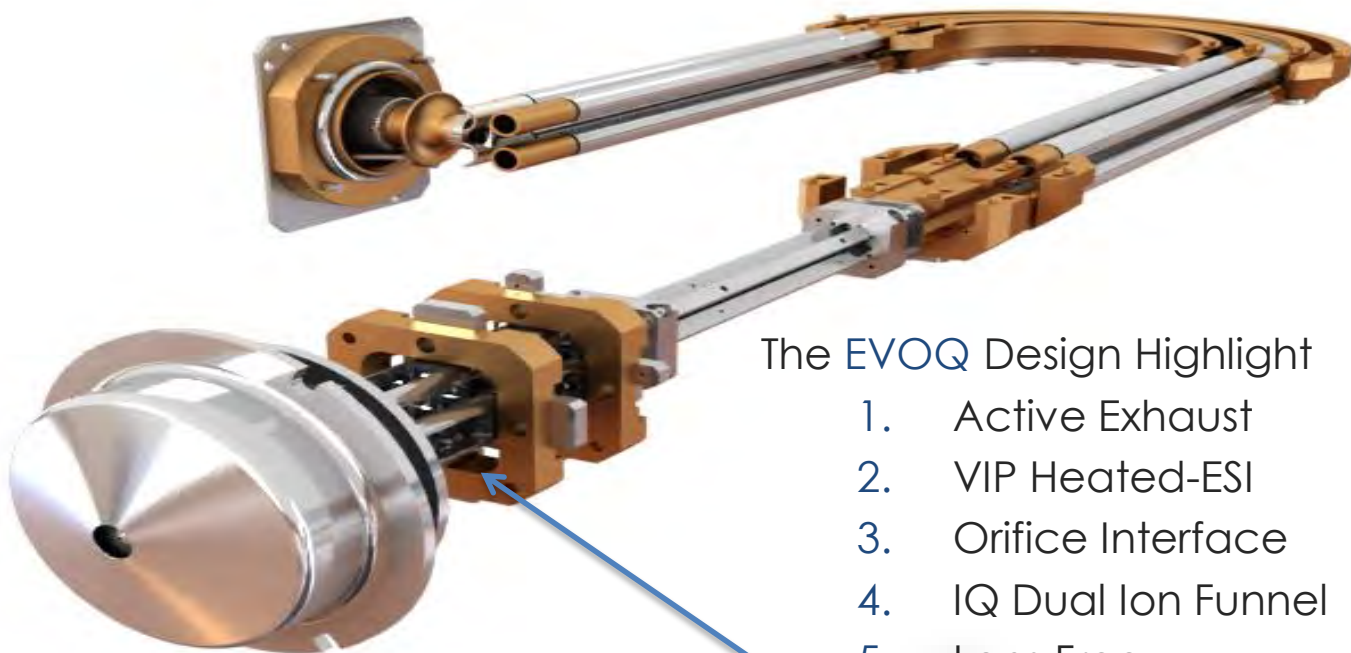
Analysis



MS Design Highlight

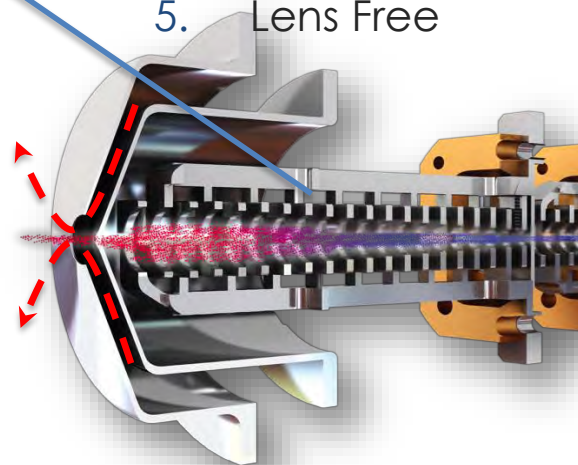


Low pressure region pulls all exhaust gases through the large exhaust opening and out of the mass spec.



The EVOQ Design Highlight

1. Active Exhaust
2. VIP Heated-ESI
3. Orifice Interface
4. IQ Dual Ion Funnel
5. Lens Free



LC/MS Conditions

- Trap Column: YMC-Pack ODS-AQ, 10 μm , 10 mm x 3.0 mm I.D.
- Mobile Phase C: 5 mM ammonium fluoride in water
- Equilibration flow: 1000 $\mu\text{L}/\text{min}$ (3.0 min)
- Loading Flow: 600 $\mu\text{L}/\text{min}$
- Analytical Column: YMC-Pack ODS AQ, 3 μm , 150 mm \times 3.0 mm (I.D.)
- Column Temperature: 40 $^{\circ}\text{C}$
- Injection Volume: 50 μL
- Mobile Phase A: 5 mM ammonium fluoride in water
- Mobile Phase B: MeOH

LC Gradient:				MS Source Parameters	
Time min.	Mobile Phase A (%)	Mobile Phase B (%)	Flow Rate $\mu\text{L}/\text{min}$.	Source:	HESI
0.0	90	10	400	Spray Voltage (+/-)	4000 V
0.2	90	10	400	Cone Gas Flow	20
2.0	30	70	400	Cone Temperature	250 $^{\circ}\text{C}$
6.5	20	80	400	Heated Probe Gas Flow	45
8.0	0	100	400	Heated Probe Temperature	400 $^{\circ}\text{C}$
15.0	0	100	400	Nebulizer Gas Flow	65
15.1	90	10	400	Exhaust Gas	On
18.0	90	10	400		

MRM Transitions

	Name	Retention Time	RT Window	CAS Number	Retention Index	Scan Type	Scan Time (ms)	Polarity
1	2,3,5_trimethacarb	7.39	1.00		0	MRM	24.7	Positive
2	2,4-D	6.24	1.00		0	MRM	24.7	Negative
3	2,6 dichlorbenzamide	4.86	1.00		0	MRM	23.0	Positive
4	3-Hydroxycarbofuran	5.05	1.00	16655-82-6	0	MRM	23.0	Positive
5	Abamectin	13.52	1.00	71751-41-2	0	MRM	47.6	Positive
6	Acephate	4.16	1.00	30560-19-1	0	MRM	23.8	Positive
7	Acetamiprid	5.10	1.00	135410-20-7	0	MRM	23.0	Positive
8	Aldicarb sulfone	4.40	1.00	1646-88-4	0	MRM	23.0	Positive
9	Aldicarb sulfoxide	4.28	1.00	1646-87-3	0	MRM	23.0	Positive
10	Ametryn	8.52	1.00	834-12-8	0	MRM	19.6	Positive
11	Aminocarb	6.33	1.00	2032-59-9	0	MRM	24.7	Positive
12	Atrazine	7.39	1.00		0	MRM	24.7	Positive
13	Azoxystrobin	7.74	1.00	131860-33-8	0	MRM	23.0	Positive
14	Benalaxyl	11.14	1.00	71626-11-4	0	MRM	24.7	Positive
15	Bendiocarb	6.11	1.00	22781-23-3	0	MRM	24.7	Positive
16	Benfuracarb	12.27	1.00	82560-54-1	0	MRM	23.8	Positive
17	Bentazone	5.02	1.00		0	MRM	23.0	Negative
18	Bifenazate	8.90	1.00	149877-41-8	0	MRM	19.6	Positive
19	Bitertanol	11.42	1.00	55179-31-2	0	MRM	23.8	Positive
20	Boscalid	8.32	1.00	188425-85-6	0	MRM	19.6	Positive
21	Bromucanozole Isomer 1	9.35	1.00	116255-48-2	0	MRM	20.8	Positive
22	Bromucanozole Isomer 2	10.69	1.00	116255-48-2	0	MRM	30.3	Positive
23	Bupirimate	10.42	1.00	41483-43-6	0	MRM	30.3	Positive
24	Buprofezin	12.48	1.00	69327-76-0	0	MRM	23.8	Positive
25	Butafenacil	8.96	1.00	134605-64-4	0	MRM	19.6	Positive

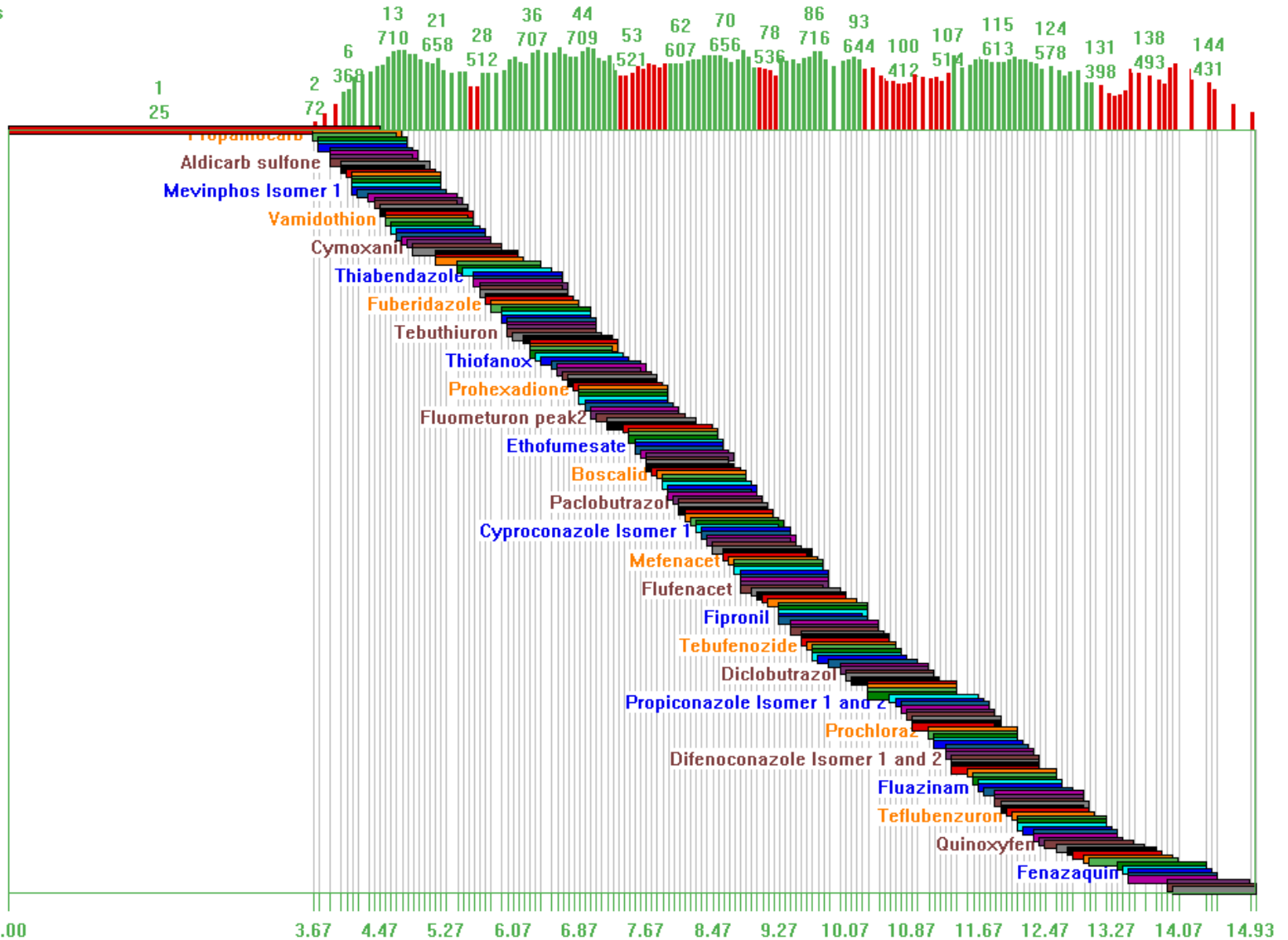
	Precursor	Product	Collision Energy	Q1 Resolution	Q3 Resolution	Scan Time (%)	Qualifier Ion	Qualifier Ratio	Quantifier Ion
1	194.00	121.90	24.00	Standard (2.0)	Standard (2.0)	50.00%	<input checked="" type="checkbox"/>	50.20%	<input type="checkbox"/>
2	211.00	137.00	13.00	Standard (2.0)	Standard (2.0)	50.00%	<input type="checkbox"/>		<input checked="" type="checkbox"/>
3							<input type="checkbox"/>		<input type="checkbox"/>
4							<input type="checkbox"/>		<input type="checkbox"/>
5							<input type="checkbox"/>		<input type="checkbox"/>
6							<input type="checkbox"/>		<input type="checkbox"/>
7							<input type="checkbox"/>		<input type="checkbox"/>

Timed MRM Windows for 215 Pesticides (430 MRM Transitions)

215 Compounds

Scan Times (ms):

Retention Times (min): 0.00

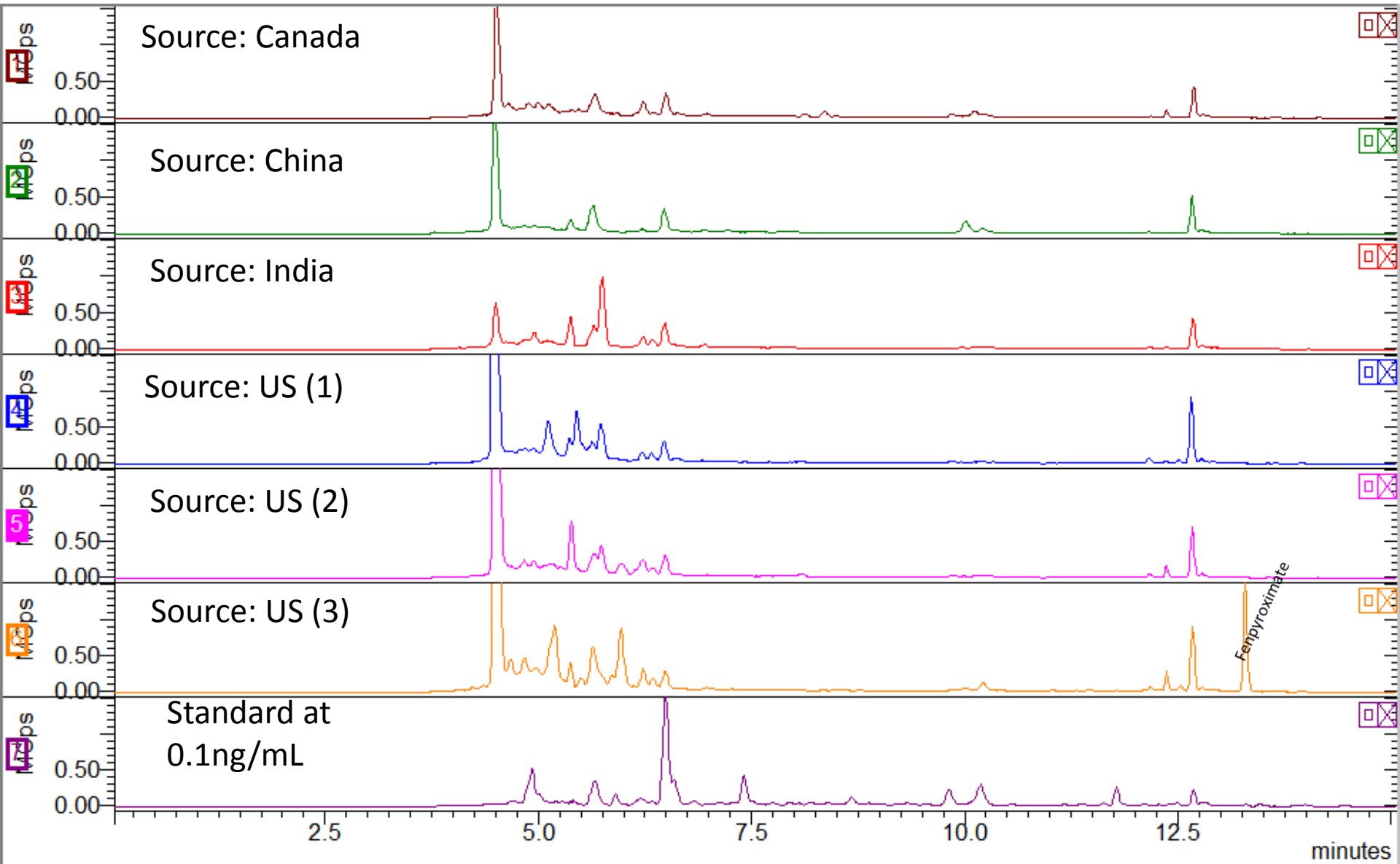


Result

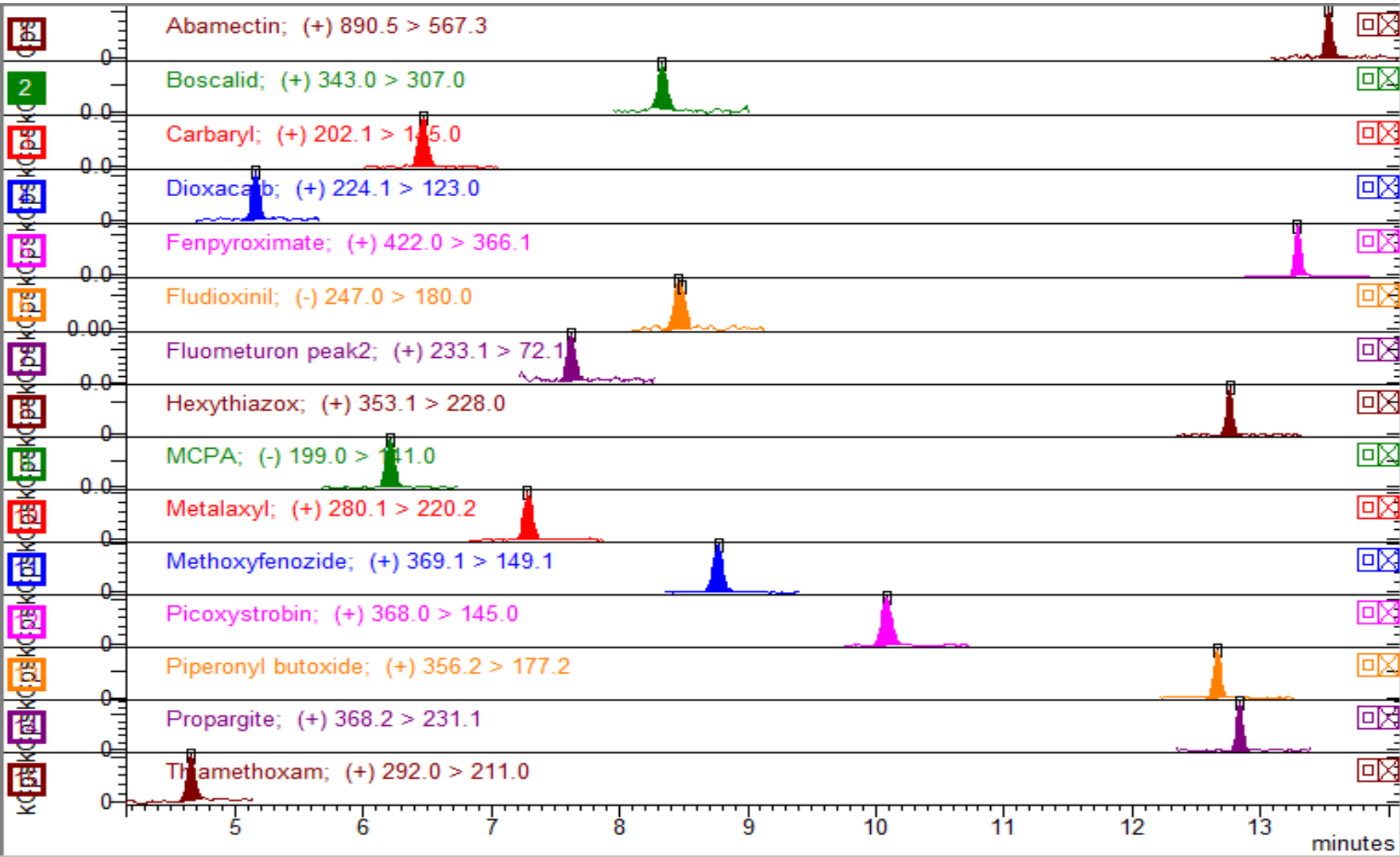
Honey Source=>	India	Canada	China	US-1	US-2	US-3
Pesticide		ng/g (ppb)				
Acetamiprid	ND	ND	0.6	ND	ND	ND
Boscalid	ND	17.5	ND	ND	0.15	3.4
Carbaryl	ND	0.71	ND	ND	ND	ND
Dioxacarb	ND	ND	ND	ND	1.35	2
Fenpyroximate	ND	ND	ND	ND	0.26	55
Fludioxinil	ND	1.5	ND	ND	ND	ND
Fluometuron	ND	ND	ND	ND	ND	2.8
Hexythiazox	ND	ND	0.2	ND	ND	ND
MCPA	ND	0.7	ND	ND	ND	ND
Metalaxyl	ND	0.1	ND	ND	ND	ND
Methoxyfenozide	ND	ND	ND	ND	ND	0.9
Picoxystrobin	ND	4.23	ND	ND	ND	ND
Piperonyl butoxide	ND	0.3	ND	0.6	0.8	0.2
Propargite	ND	0.3	ND	0.1	ND	ND
Thiamethoxam	ND	4.9	ND	ND	ND	ND

Test result (ND= not detected or <0.1 ppb)

Honey TIC Chromatograms

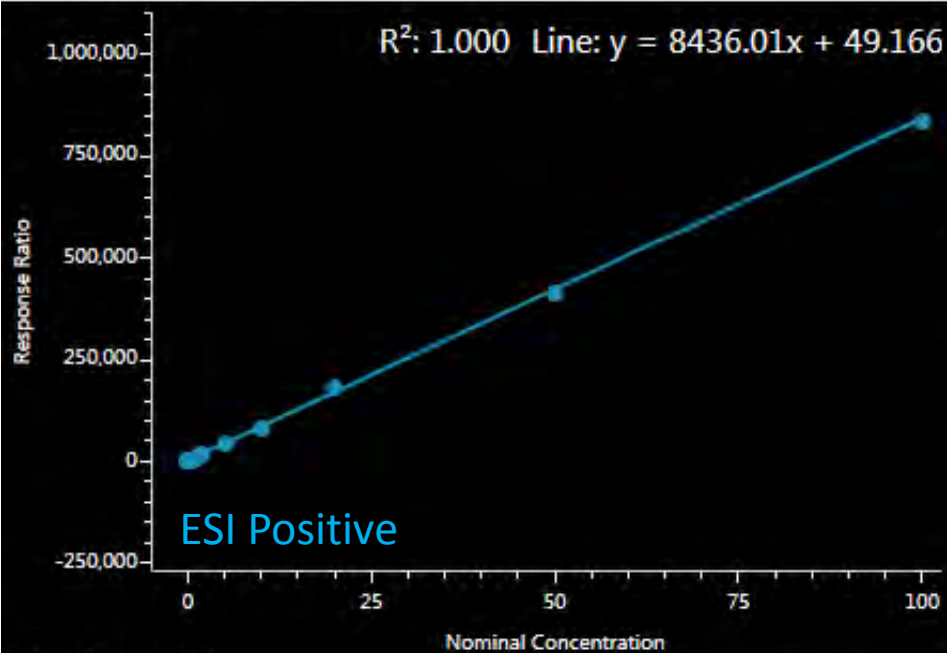


Pesticides at 0.05 ng/mL

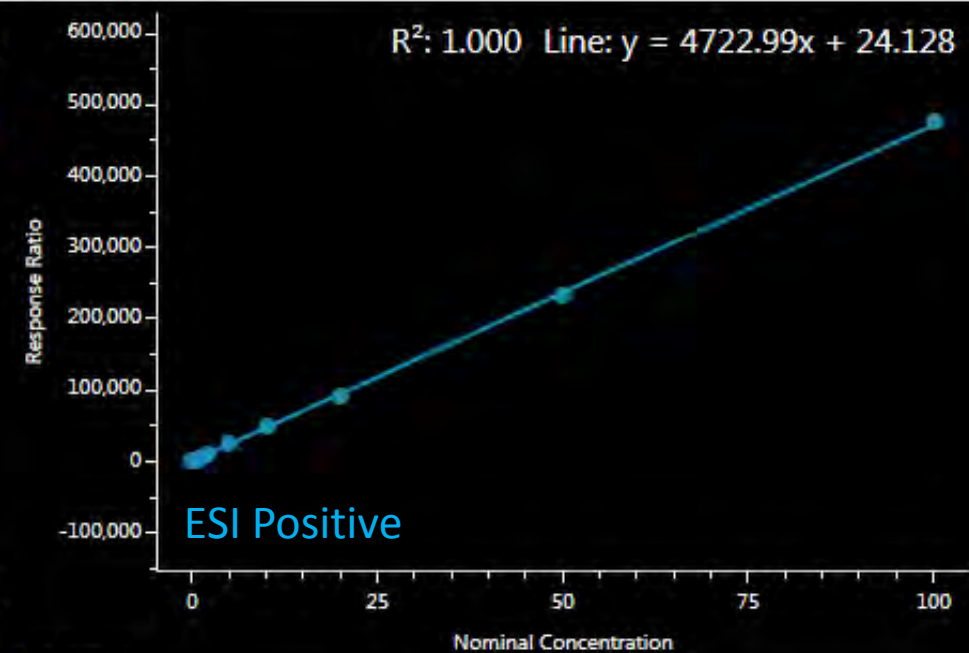


Above pesticides listed in result table

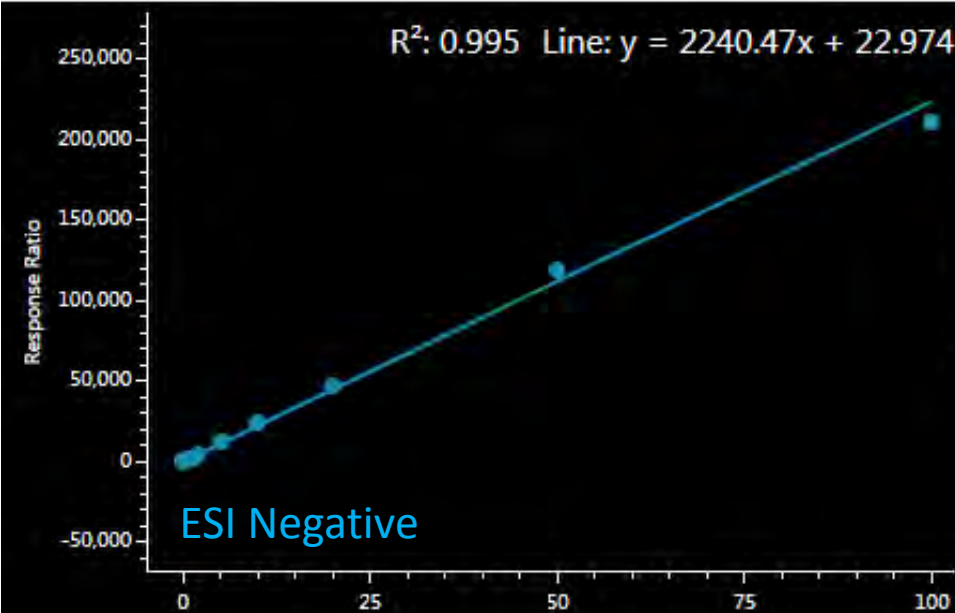
Acetamiprid



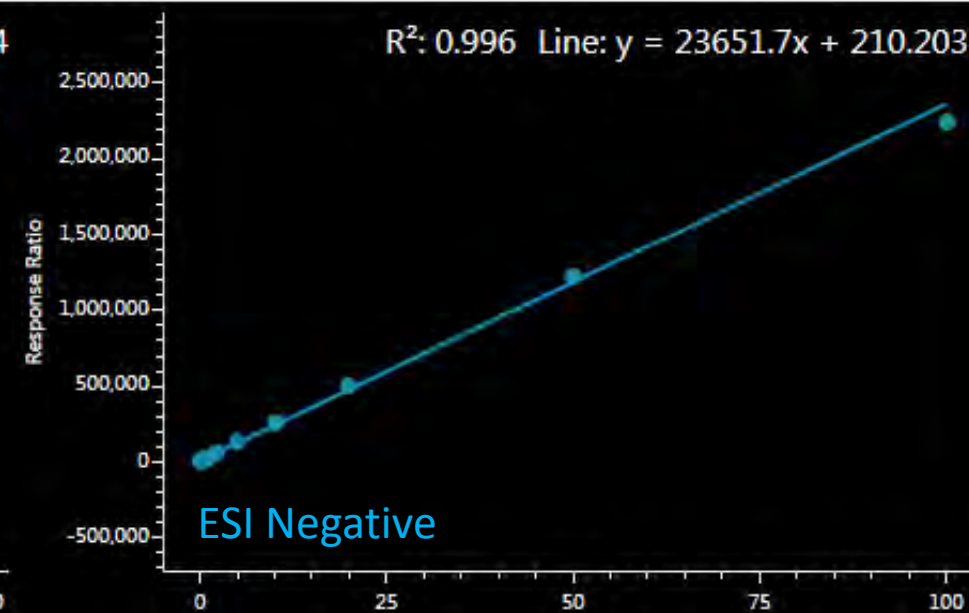
Dioxacarb



Fludioxinil



MCPA





Peak Review

Fenpyroximate

Determination Review

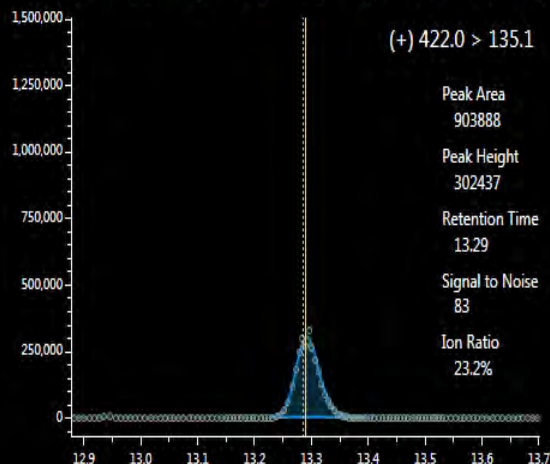
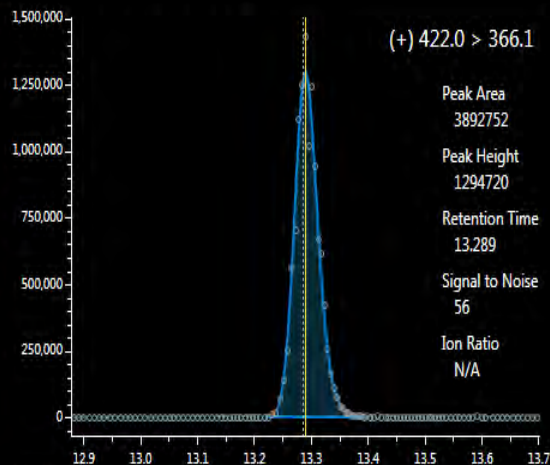
Honey_NN3

1 of 1 Samples

Fenpyroximate

204 of 215 Compounds

Determination Chromatograms



Determination Calculation

Concentration 54.688
Response 3892752

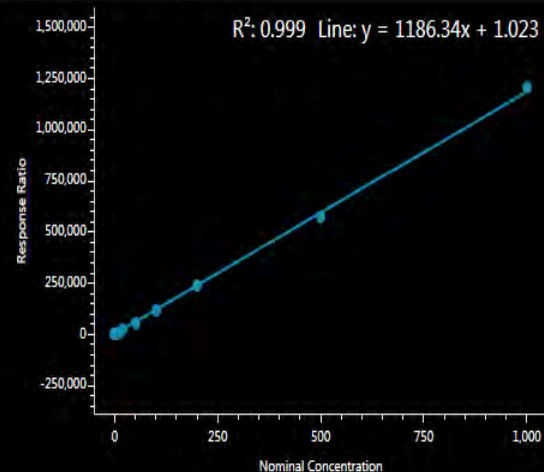
Determination Flags

There are no flags for this determination.

Determination Information

Sample Honey_NN3
Vial TR: CStk1-01 VL: 21
Injection Number 0
Sample Type Unknown
Calibration Level 1
Dilution 1

Determination Calibration



Summary

- The sample prep step is quick, simple, and easy of use.
- The procedure provides a more convenient and simpler approach than QuEChERS and SPE for residue chemical analysis in honey.

Acknowledgment

THOMSON INSTRUMENT COMPANY

