# Screening and Quantitation of 250 Pesticides in Fruit Juices with Positive/Negative Switching LC/MS/MS

#### Florida Pesticide Residue Workshop 2014

**Zicheng Yang and Louis Maljers** 

Bruker, Chemical & Applied Markets (CAM) Division, 3500 W Warren Ave, Fremont, CA USA 94538

Contact: zicheng.yang@bruker.com

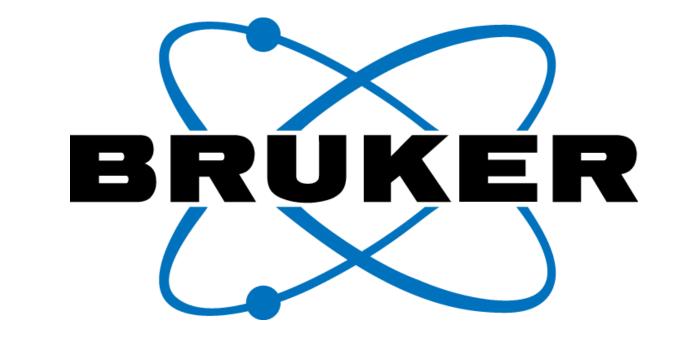
# **Sample Preparation**

Mix 50  $\mu$ L fruit juice with 450  $\mu$ L of solvent (MeOH/water, 10/90, v/v) in the filter vial (Part number 85531-5, Thomson Instrument Company) and press filter plunger (0.2  $\mu$ m PVDF) to filter.

## Introduction

### Methods

		Orange	Cranberry	White Grape	Vegetable	
Fruit Juice=>	Apple Juice	Juice	Juice	Juice	Juice	
Pesticide			μg/L (pp	b)		
Azoxystrobin	ND	ND	0.32	ND	0.48	
Boscalid	ND	ND	0.16	ND	ND	
Carbaryl	ND	0.39	1.47	ND	ND	
Carbofuran	ND	0.14	ND	ND	ND	
Dimethoate	ND	0.30	ND	ND	ND	
Imidacloprid	ND	ND	0.60	ND	0.20	
Mandipropamid	ND	ND	0.59	ND	ND	
Metalaxyl	ND	ND	0.21	ND	ND	



Liquid chromatography coupled with tandem mass spectrometry operated in multiple reaction monitoring (MRM) mode with electrospray ionization (ESI) is widely used for polar, semivolatile, and thermally labile pesticides in food testing. Many contract labs currently perform multi-residue analysis of pesticides using separate positive and negative methods due to instrument limitations especially for methods with hundreds of MRM transitions. This requires twice the sample and twice the analysis time. Recently, the Bruker EVOQ Elite LC-triple quadrupole system has been introduced to the market; thereby providing fast positive/negative switching allowing for simultaneous determination of positive and negative co-eluting compounds numbering in the hundreds.

#### Instruments:

EVOQ Elite triple quadrupole mass spectrometer coupled to a Bruker UHPLC and CTC Autosampler (see Fig. 1)

#### LC Parameters:

Column: YMC-Pack ODS-AQ 3 µm, 150mm x 3mm (I.D.) Column Temperature: 40 °C Injection Volume: 30 µL Mobile Phase A: 5 mM ammonium Fluoride in water Mobile Phase B: Methanol Gradient:

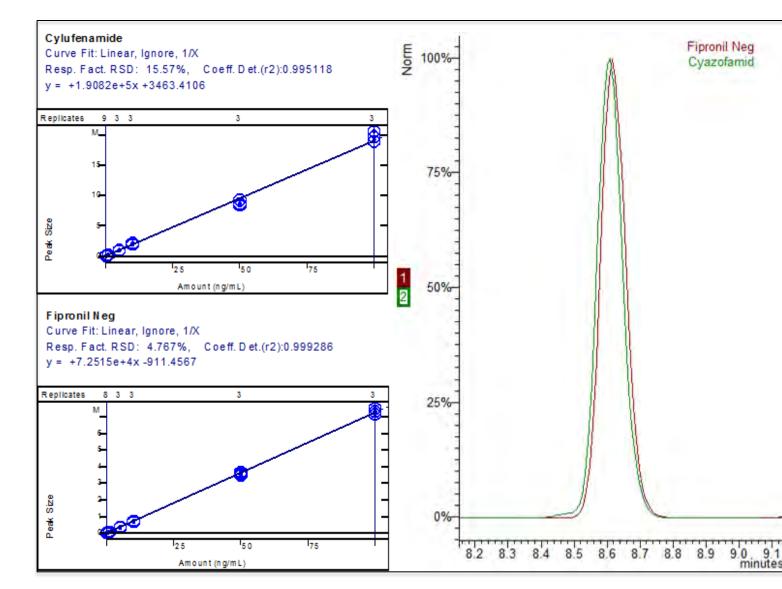
Time	%A	%B	Flow
			(µL/min
0.00	90	10	400
0.20	90	10	400
2.00	30	70	400
10.0	0	100	400
15.0	0	100	400
15.1	90	10	500
17.5	90	10	500
18.0	90	10	400

Methoxyfenozide	ND	ND	ND	ND	0.84
Tebuconazole	ND	ND	0.32	ND	ND
Thiabendazole	1.8	ND	ND	ND	ND

#### Table 1. Test result (ND= not detected or <0.1ppb)

	4		Nar	me		Retentio Time	Ŋ	RT Window	CAS	Number		tention Index	S	can Type	Scan Tim	ie (ms)	Polarity	
13	39	Mefen	acet			8.1	12	0.90				0	MRM	1		16	Positive	
14	40	Paclita	xel			8.1	14	0.90		0 1		MRM	1		16 Pos			
14	41	Tetrac	onazole			8.1	15	0.90				0	MRM	1		16	Positive	
14	42	lprova	licarb Isome	er 1 and 2		8.1	18	0.90				0	MRM	I		16	Positive	
14	43	Cyproconazole Isomer 2			8.19		0.90		0		0	MRM		16		Positive		
14	44	Flufen	acet			8.2	24	0.90				0	MRM	1		16	Positive	
14	45	Fenhe	xamid			8.2	26	0.90			0 MRM		1	17		Positive		
14	46	Spirote	etramat			8.2	29	0.90				0	MRM	1		17	Positive	
14	47	Bromu	canozole Is	somer 1		8.2	29	0.90				0	MRM	1		17	Positive	
14	48	Fluquir	nconazole			8.3	31	0.90				0	MRM		17		Positive	
14	49	Tritico	nazole			8.4	43	0.90				0	MRM		17		Positive	
15	50	Fenari	mol			8.5	51	0.90				0 MRM		17		Positive		
15	51	Cyazo	famid			8.6	53	0.90				0	MRM	1		19	Positive	
15	52	Fipron	il Neg			8.6	53	0.90				0	MRM	1		19	Negative	
15	53	Mepan	ipyrim			8.6	54	0.90				0	MRM			19	Positive	
15	54	Prome	tryne			8.6	65	0.90				0 MRM		19		Positive		
15	55	Fenbu	conazole			8.6	67	0.90				0	MRM			19	Positive	
15	56	Epoxic	onazole			8.7	77	0.90	0.90			0	0 MRM		19		Positive	
15	57	Etacor	nazole Isom	er 1 and 2		8.7	79	0.90				0	MRM	1		19	Positive	
15	58	Cletho	dim Isomer :	2		8.8	31	0.90				0 MRM			19	Positive		
15	59	Fluben	idiamide			8.8	36	0.90				0 MRM		19		Positive		
16	60	Spirox	amine Isom	er 1 and 2		8.8	37	0.90				0 MRM		19		Positive		
16	61	Terbut	iryn			8.9	91	0.90				0	MRM	1		19	Positive	
16	62	Flusila	zole			8.9	91	0.90				0	MRM	1		19	Positive	
16	63	Diflube	enzuron			8.9	96	0.90				0	MRM	1		19	Positive	-
	Precu	ursor	Product	Collision Energy	Q1	Resolutio	n	Q3 Resolu	ition	Custom F	Res	Scan Ti (%)		Qualifier Ion	Qualifier Ratio	Quant Ion		
1	435	.00	330.00	20.00	Unit	(0.7)	T	Custom	-	1	.00	50.	00%	<b>V</b>	123.109			
2	435	.00	250.00	20.00	Unit	(0.7)	V	Custom	-	1	.00	50.	00%			1		
3							T		-									
4							•		-									
5									-									
6							•		-									
7							T		-								-	

#### Fig. 3. MRM method for pesticides



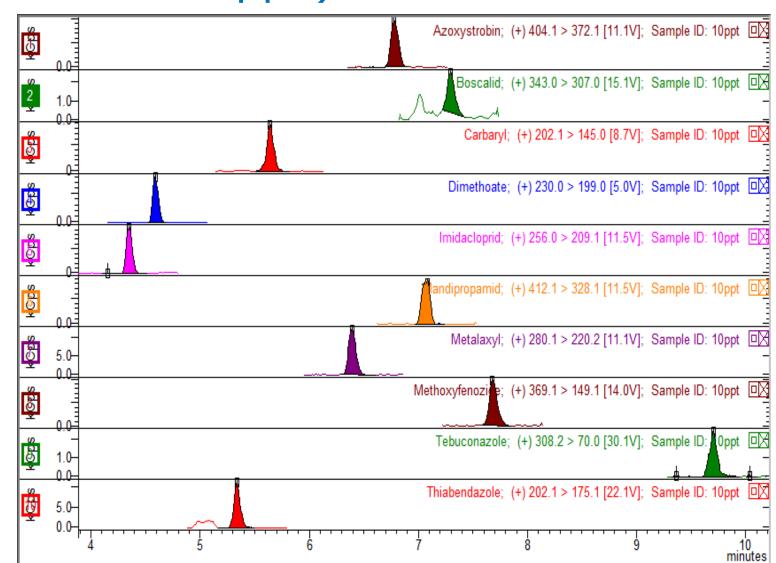


Fig. 6. Chromatograms of standard solution of the compounds listed in table 1 at 0.01 ppb (equivalent to 0.1ppb in juice).

The calibration on triplicate injections showed excellent linearity and response factor RSD over 3 orders range (Fig 4.).

A study using the EVOQ analyzed 250 pesticides in apple juice, cranberry juice, grape juice, orange juice and vegetable juice using only one method with positive negative switching for over 500 MRM transitions. The measurements were conducted by dilute-and-shoot without sample enrichment. The fruit juices were diluted 10-fold and filtered by filter vial prior to injection. An YMC-Pack ODS-AQ, 3 µm, 150 mm x 3 mm (I.D.) column with mobile phases (A) 5 mM ammonium fluoride in water, and (B) methanol were used. The total run time was 18 minutes including reequilibration.

MS Parameters: Source: HESI Spray Voltage (Positive):4000V Spray Voltage (Negative):4000V Cone Gas Flow:20-unit Cone Temperature:250° C Heated Probe Gas Flow:40-unit Heated Probe Temperature:400°C Nebulizer Gas Flow:60-unit Exhaust Gas:On

# **Results & Discussion**

The compound based scanning (CBS) can automatically compute and assign the scan (dwell) time for each MRM (Fig. 2) for timed MRM, based on peak width and Fig. 4. Calibration curve of negative pesticide Fipronil (left top) and positive pesticide Cyazofamid (left bottom), and their co-eluting plots (right).

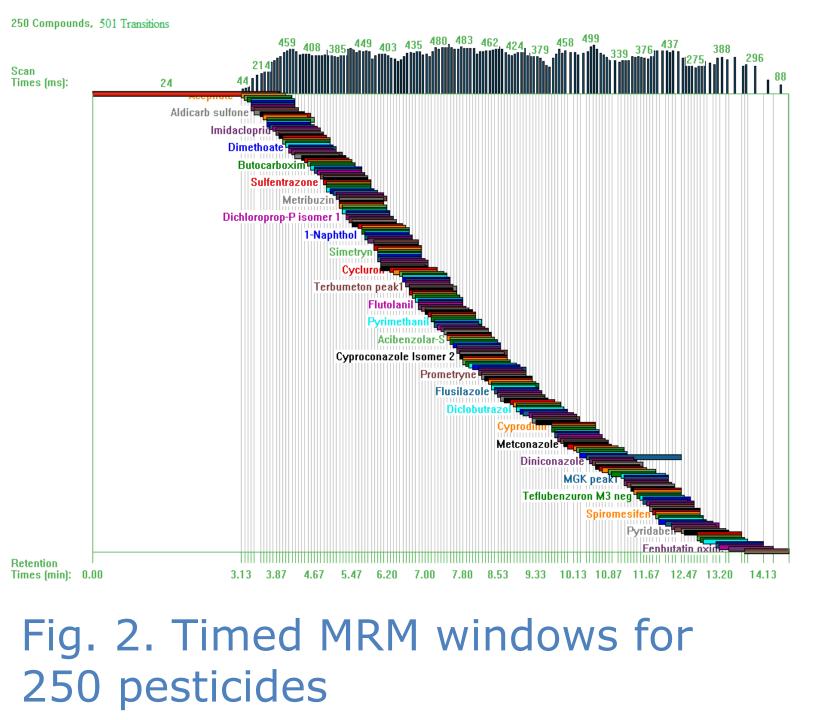
Compound No. 151 (positive Cyazofamid) and No. 152 (negative Fipronil) have same retention time and assigned same scan time (Fig 3).

Single injection of standard calibration solutions were injected before juice samples and duplicate injections of standard calibration solutions were

- Three orders Linear range:0.01ppb to 10ppb (equivalent to 0.1 ppb to 100 ppb in fruit juice).
- R2 0.999 for both pesticides.
- Response factor RSD<5%
- A total of twelve pesticides were detected in apple juice, orange juice, cranberry juice or vegetable juice (table 1.).
- There were no pesticides detected in white grape juice using the screening method (table 1.).
- The multiple pesticides detected in orange juice, cranberry juice and juice may suggest that juice comes from multiple sources of raw materials or pooled juices.

Fig. 1 EVOQ Elite triple quadrupole mass spectrometer coupled to a Bruker UHPLC and CTC Autosampler

#### data points required.



#### performed after the sample.

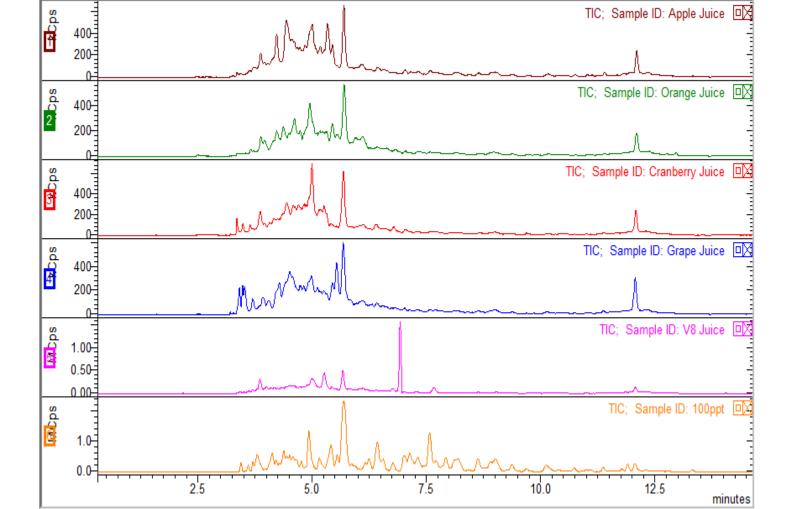


Fig. 5. Total Ion Chromatograms of (top to bottom): Apple juice, orange juice, cranberry juice, grape juice and vegetable juice.

# Conclusions

 Good linearity, sensitivity and response factor RSD for positive and negative coeluting pesticides.

 Bruker EVOQ Elite LC/MS/MS is a system of choice for simultaneous determination of positive and negative compounds numbering in the hundreds.

#### TIC-PL-082-276