Agilent QuEChERS Kits

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Product Manager SPE

EPRW 2010

What is QuEChERS (pronounced "*Catchers*")

- Quick, Easy, Cheap, Effective, Robust and Safe
- Sample Preparation method developed jointly by US FDA and EU Food Regulatory Agencies
- Methodology for simplified extraction of a large number of pesticide residues in fruits and vegetables
- QuEChERS is a fast sample preparation method for screening for analytes of interest, not for concentrating samples
- QuEChERS is still in it's "Infancy"; being adopted worldwide

	Luke method or traditional SPE	QuEChERS	Bottom Line
Estimated Time to process 6 samples (min)	120	30	4x faster
Solvent Used (mL)	90 mL	10mL	9 x less solvent
Chlorinated Waste (mL)	30 mL	none	safer, greener, less costly
Glassware/ specialized equipment	Funnel, water bath, 200mL containers, evaporator, etc.	None	No additional supplies needed

QuEChERS Brochure: literature #5990-3562EN

Comparison of the 3 Methods

- Original QuEChERS method (unbuffered)
  - 4 or 6 g MgSO4, 1 or 1.5 g NaCl
- AOAC method 2007.01 (AOAC)
  - 6 g MgSO4, 1.5 g Na Acetate
- EN method 15662 (CEN)
  - 4 g MgSO4, 1 g NaCl, 1 g NaCitrate, 0.5 g disodium citrate sesquihydrate

All 3 methods give excellent results: average 98% recoveries with 10% RSDs

Ionic Strength of the two methods:

- AOAC buffered method
  - Relatively strong acetate buffering conditions (pH 4.8)
- EN buffered method
  - Weaker citrate buffering conditions (pH 5-5.5)

Universal d-SPE

- 50 mg PSA, 50 mg C18, 7.5 mg GCB, 150 mg MgSO4

- Cleanest extract, for all matrices without unacceptably affecting recoveries even for structurally planar pesticides

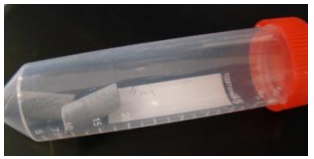
J. Chromatography A 1217 (2010) 2548-2560.

Extraction Choice

- AOAC over EN over Unbuffered

The *Value* of Agilent SampliQ QuEChERS Kits

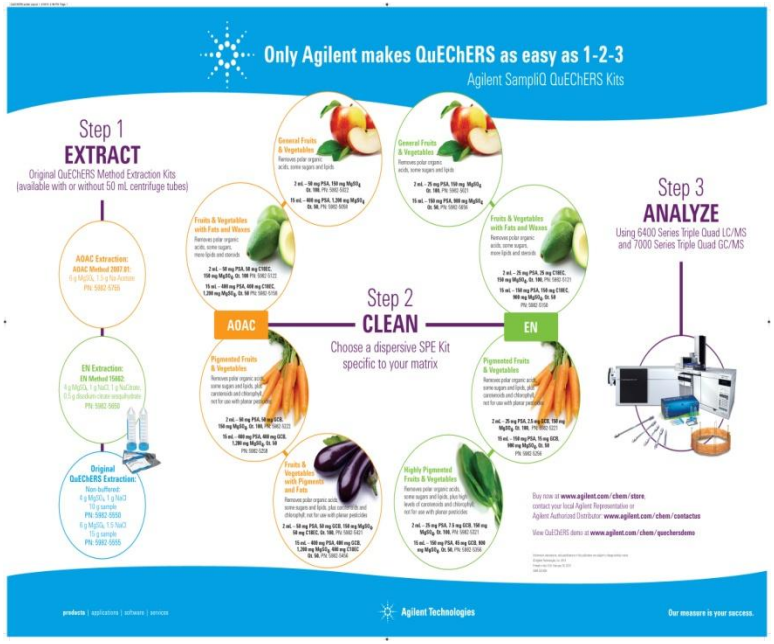
- **Reduced time and labor:** New **Ceramic Homogenizers**
  - With addition of ceramic homogenizers in tubes, shaking step is minimized to only 20 seconds
  - Saves you time, money and improves results
  - Proof of performance included in QuEChERS brochure (5990-3562EN)



- **Improved Recoveries: Individually packaged extraction salts**
  - Anhydrous foil packs ensure salt integrity, under nitrogen
  - Avoid exothermic reactions by adding salts *after* solvent
  - Exothermic reactions cause samples to degrade
  - No need for additional centrifuge tubes

- **Easy Product Selection: QuEChERS Kits Identified by method and matrix**
  - Select an extraction kit based on methodology (AOAC or EN)
  - Select a dispersive kit based on food type (meat, fruits, vegetables, etc)
  - Extraction packets available without tubes

- **Technical support**: Over 20 application notes on QuEChERS:
  - All available on our website: www.agilent.com/sampliQ
  - QuEChERS Food Application “Notebooks”: Lit number: 5990-4977EN
  - Seminars and hands-on training available worldwide



Backed by the Agilent brand you know and trust for all your instrument and column needs!

SampliQ QuEChERS Market Leader

- ✓ Total Extraction Kit Options: 16
- ✓ Total Dispersive Kit Options: 33
- ✓ Applications: 20+
- ✓ Local support (tech support specialists, product specialists and distributors throughout Europe)
- ✓Solutions expertise (GC/MS and LC/MS)
- ✓Certificate of Composition with each kit
- ✓Satisfied customers around the world!

The most complete QuEChERS solution for customers in the market today

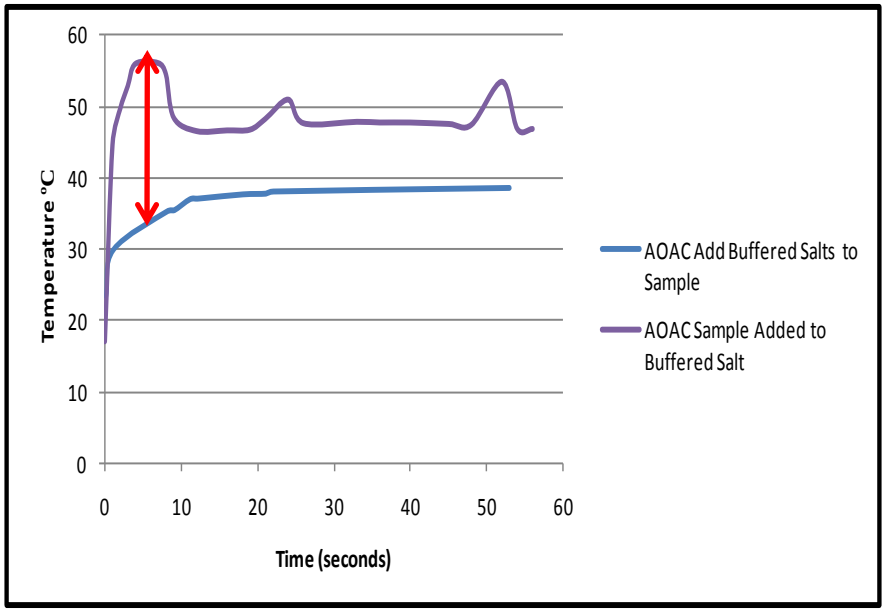
Extraction Kit Packaging

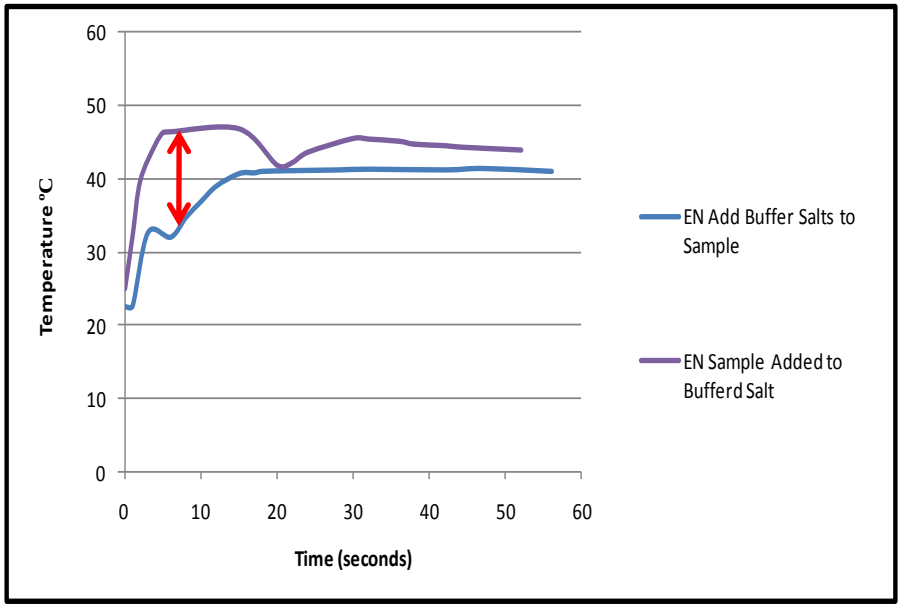
- ✓ Water and UV resistant packaging 3 layer material
- ✓ Pack the salts under nitrogen not vacuum
- ✓ Increases pouring capabilities
- ✓ Substantially reduces clumping of salts
- ✓ All ingredients listed on packet

Packaging salts separately allows customers to add sample before adding salts

- Prevents exothermic reaction
- Prevents degradation of sample
- Ensures maximum recoveries

Temperature Graphs from AOAC and EN Extraction Kits

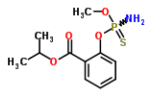




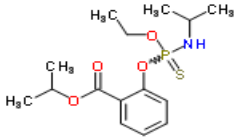
Represent the thermal data acquired from the AOAC and EN extraction kits. A substantial increase in temperature is observed (increase of approx 25 ºC for the AOAC method and approx 10 ºC for the EN method) when the produce sample is added to the extraction salt in the 50 mL PP tube versus when the extraction salt from an anhydrous pouch is added to the produce sample

Effect of Temperature on Recovery

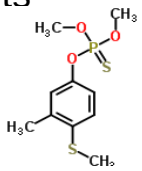
- The following four pesticides were used to evaluate the thermal effects that are prevalent when the produce is added to the extraction salts



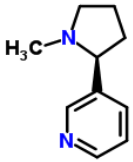
Isocarbophos



Isofenphos

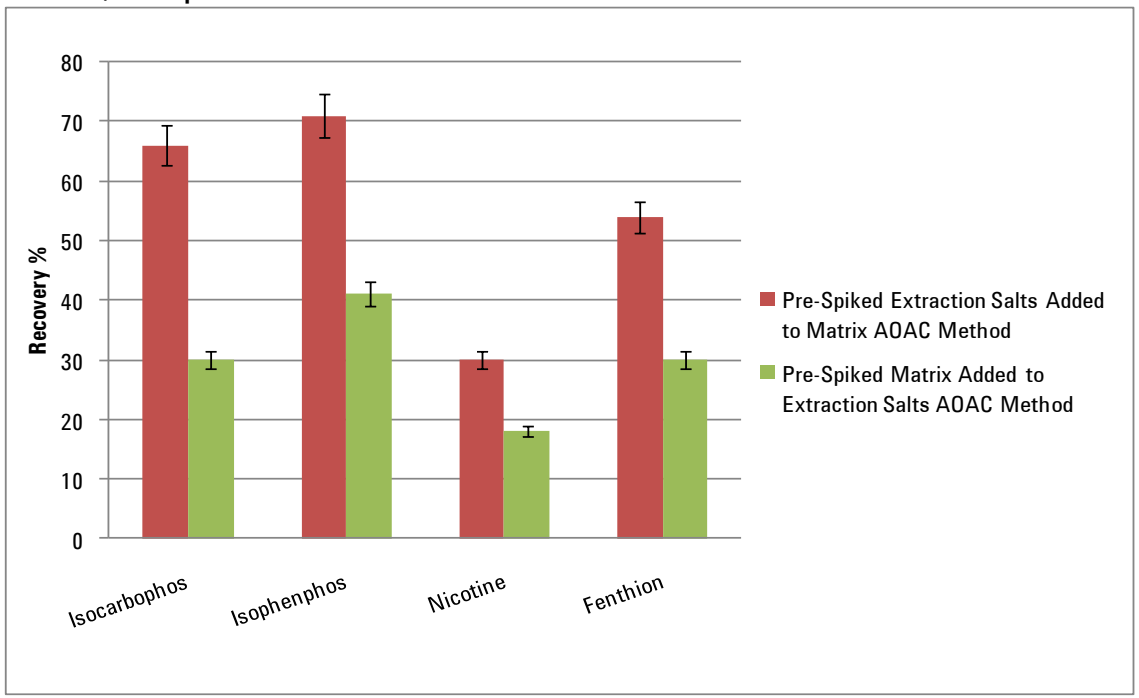


Fenthion



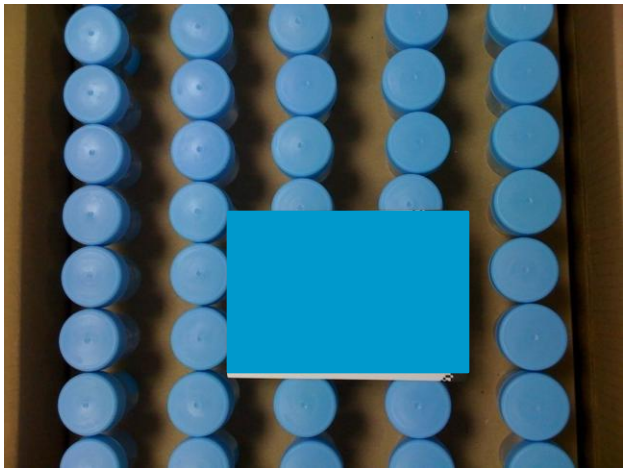
Nicotine*

- Analysis was performed on a Agilent 6410 Triple Quad LC/MS using a Phenyl- Hexyl column 2.1 x 50 mm, 3.5 µm



Represents the negative effect adding the produce matrix to the salt has on recovery during the QuEChERS extraction step, AOAC method

Other Companies' Extraction Packaging



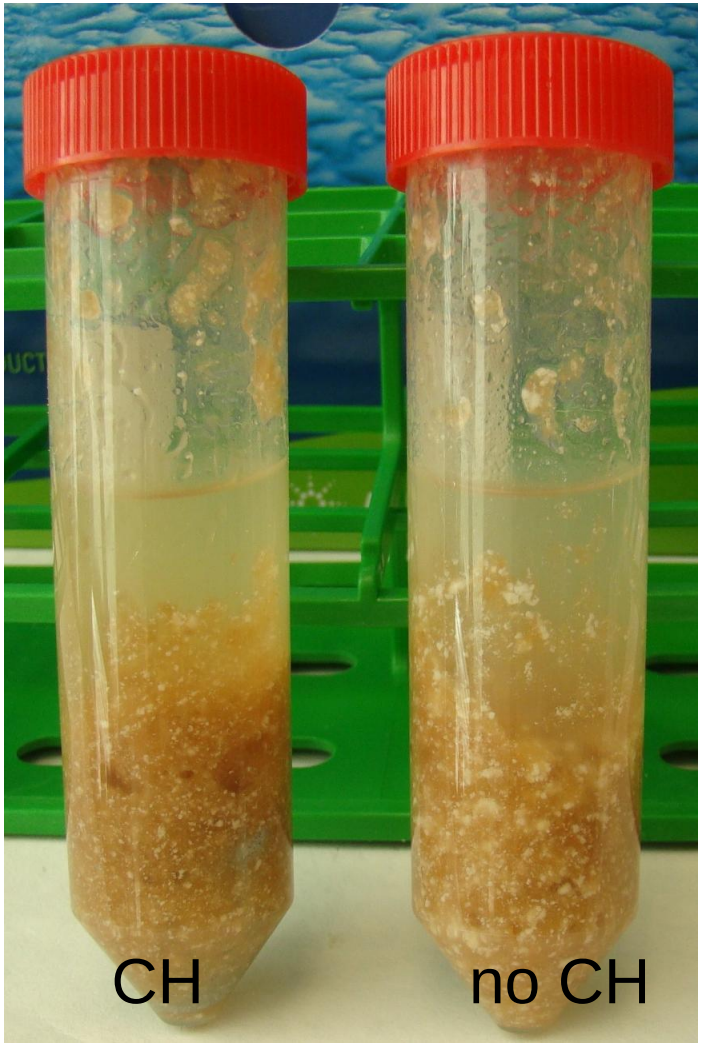
Improvements to the Extraction of Samples

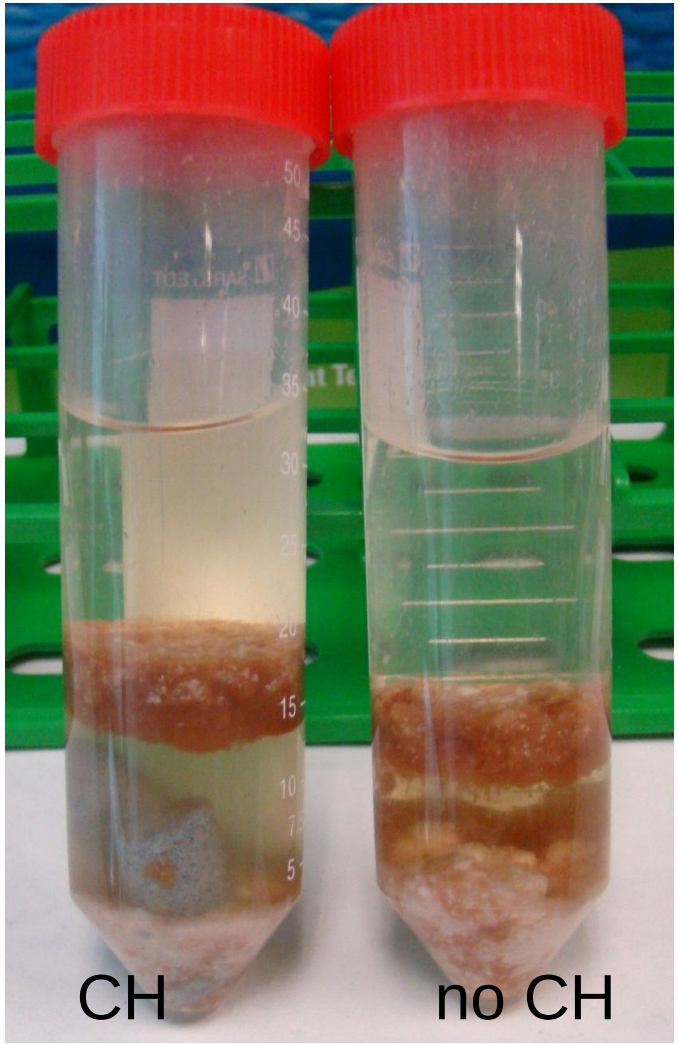
Step 1: Extraction salts with comminuted fruit/vegetable

- Consistency in shaking, everyone shakes differently
- Variability in QuEChERS applications, recovery and RSDs

- ***SampliQ Ceramic Homogenizers***
  - **Reduces shaking time from 1 minute to <20 seconds!**
  - Consistent extraction of the sample with the salts
  - Breaks up salt agglomerates
  - Facilitates homogenization with angle cut
  - Increase recovery of pesticides from sample
  - 3 different sizes: 50 mL extraction tube and 2 dispersive tubes 15 and 2 mL

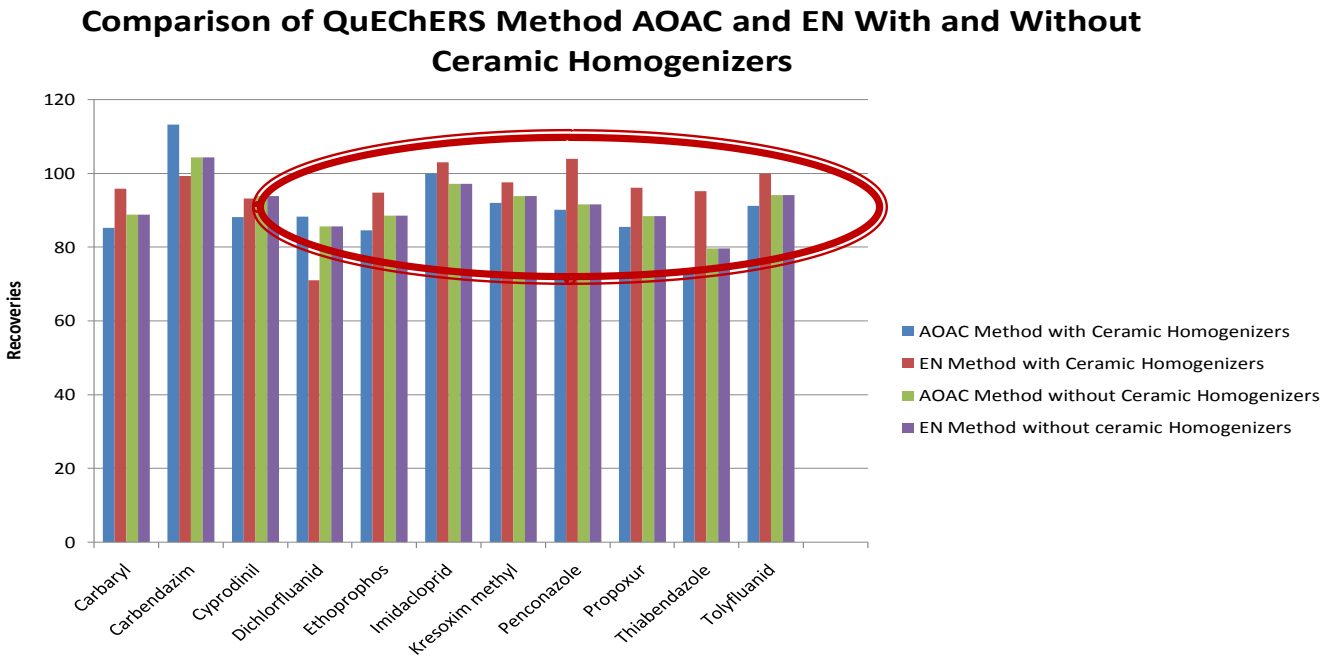
QuEChERS Extraction: With and Without Ceramic Homogenizers

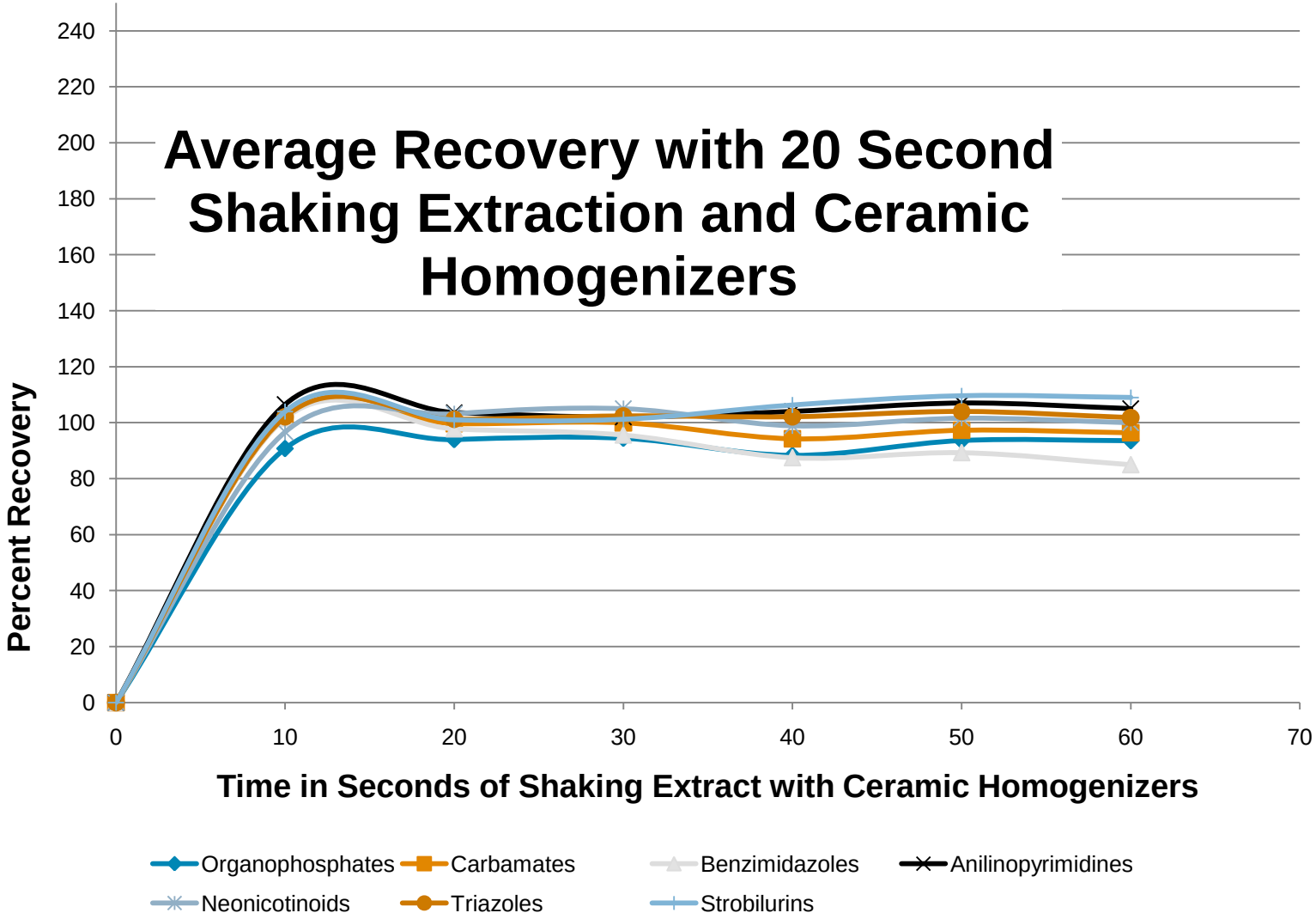




Ceramic Homogenizers

- Inert ceramic material
- No loss in pesticide recovery
- Consistent recovery





Pesticides used in study: Acephate, Carbaryl, Carbendazim, Cyprodinil, Imidacloprid, Imazalil, Methamidophos, Penconazole, Propoxur, Pymetrozine, Thiabendazole, Thiophanate-methyl, Ethoprophos, Kresoxim-methyl ; Apple matrix

QuEChERS applications in 2010: beyond just Pesticides in Fruits and Vegetables

Application	Literature	QuEChERS Kit Used
Veterinary Drugs (antibiotics) in animal tissues	5990-5085EN 5990-5086EN	5982-5650 5982-4921
PAHs in Fish	5990-5411EN	5982-5755 5982-5058
Pesticides in Olive Oil	5990-5553EN	5982-5550 5982-5122
PAH's in Soil	5990-5452EN	5982-5755 5982-5156
Acrylamides in Fried Food	5900-5940EN	5982-5850 5982-5022
Pesticides in Baby Food	5990-5028EN	5982-5755 5982-5158
PCBs in Fish and Fish Oil Supplements	Coming soon!	
Pesticides in Essential Oils	Coming soon!	
Drugs in Biologicals	Coming Soon!	

Learn more today at www.agilent.com/chem/SampliQ

Agilent's' Solutions to Customers' Concerns

Concerns	Agilent's Solution	Agilent Advantage
Salts in a tube	Salts in extraction packet	☺
Extra 50 mL tube	Empty 50 mL tubes	☺
Storage and stock of extraction and dispersive tubes	Organized box arrangement and extraction packets	☺
Shaking variability	Ceramic homogenizers	☺
Making standards: time and consistency	Standards available	☺

Appendix

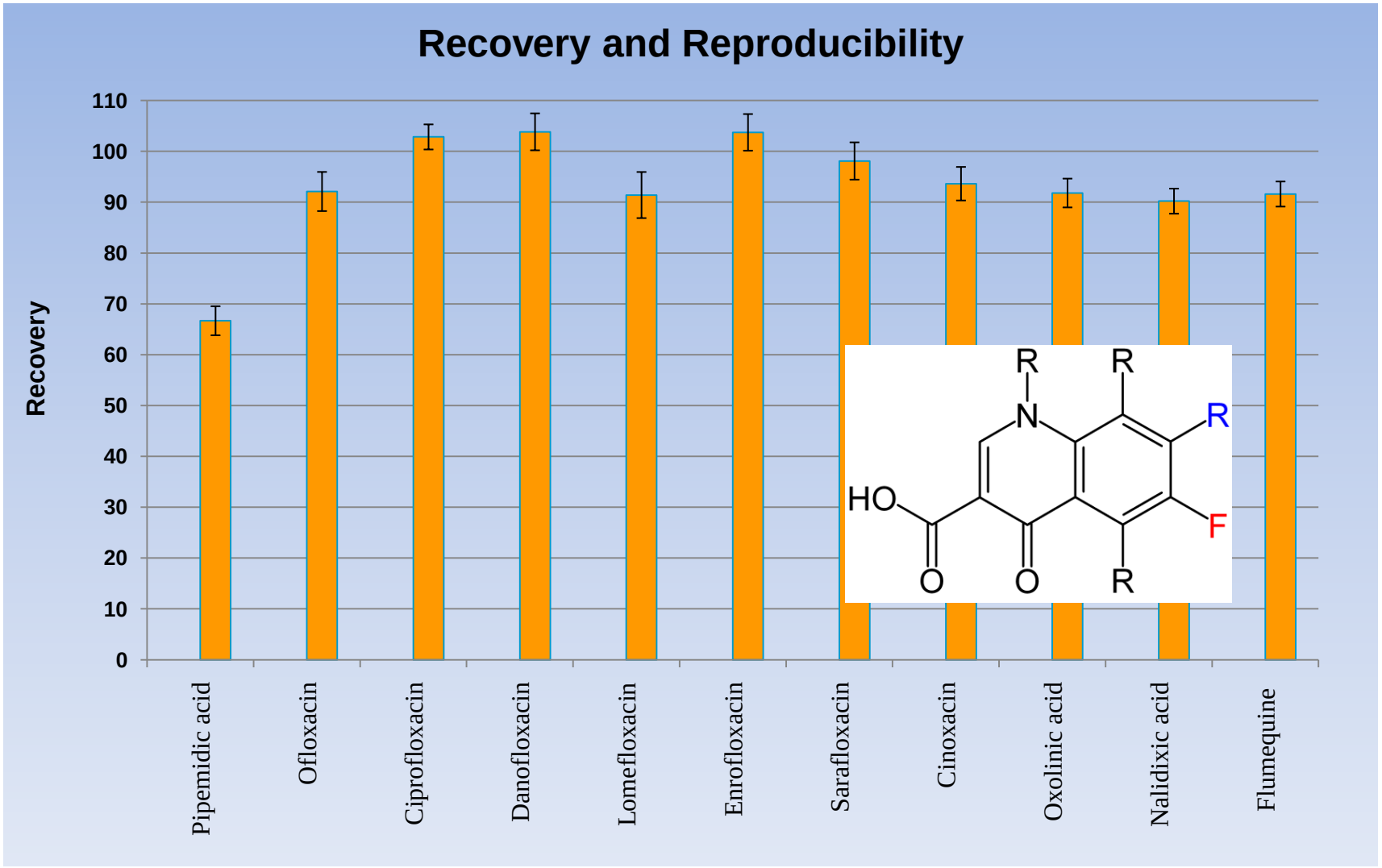
Agilent SampliQ's QuEChERS Research

- Expanding QuEChERS Methodology!
  - ✓ Pharmaceutical
    - •Veterinary Drugs in Animal Tissue
  - ✓ Environmental and Advancements in Food
    - •Pesticides in Olive Oil
    - •Pesticides in Coffee
    - •PAHs in Fish and Soil
    - •Acrylamides in Fried Foods

Cutting Edge Applications: Veterinary Drugs in Animal Tissue

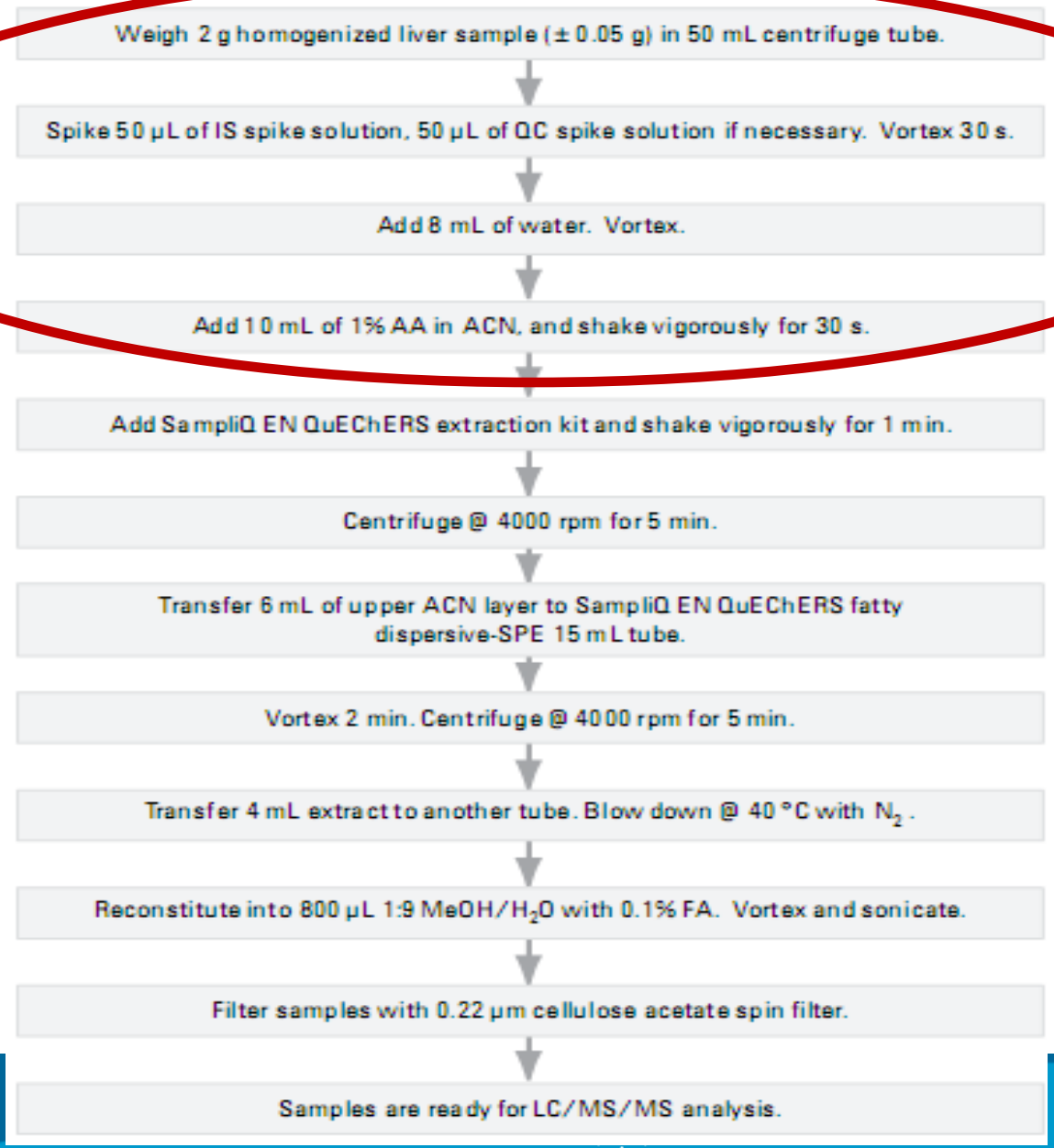
Determination of Quinolone and Sulfonamide antibiotics in bovine liver:

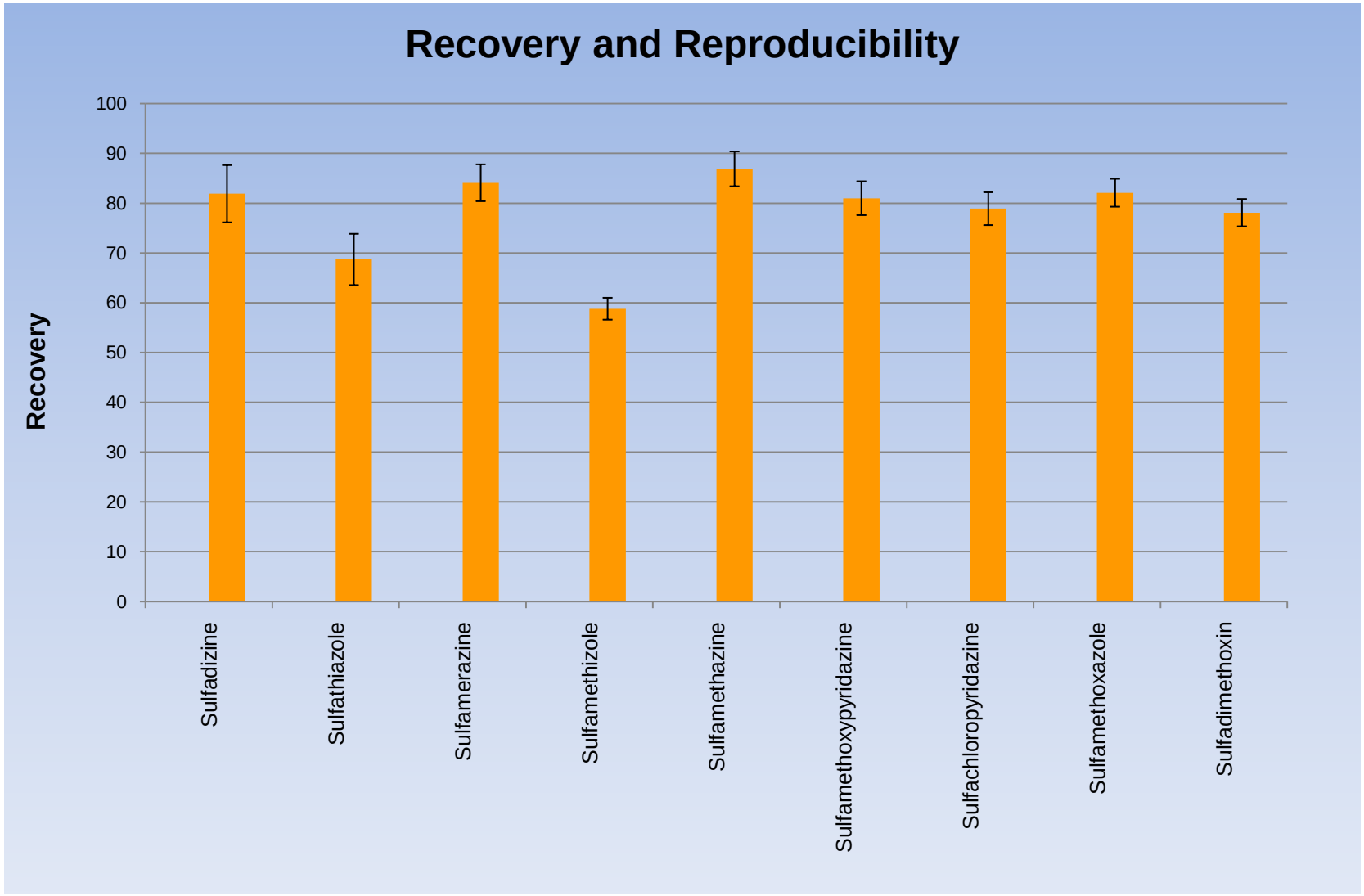
- Extraction was performed using SampliQ EN extraction kit
- SampliQ dispersive SPE kits (25 mg C18* and 150 mg $MgSO_4$), for drug residue in animal tissue **(NEW)**
- Analyzed by LC/MS/MS



Recoveries for the 11 Quinolones from Bovine Liver

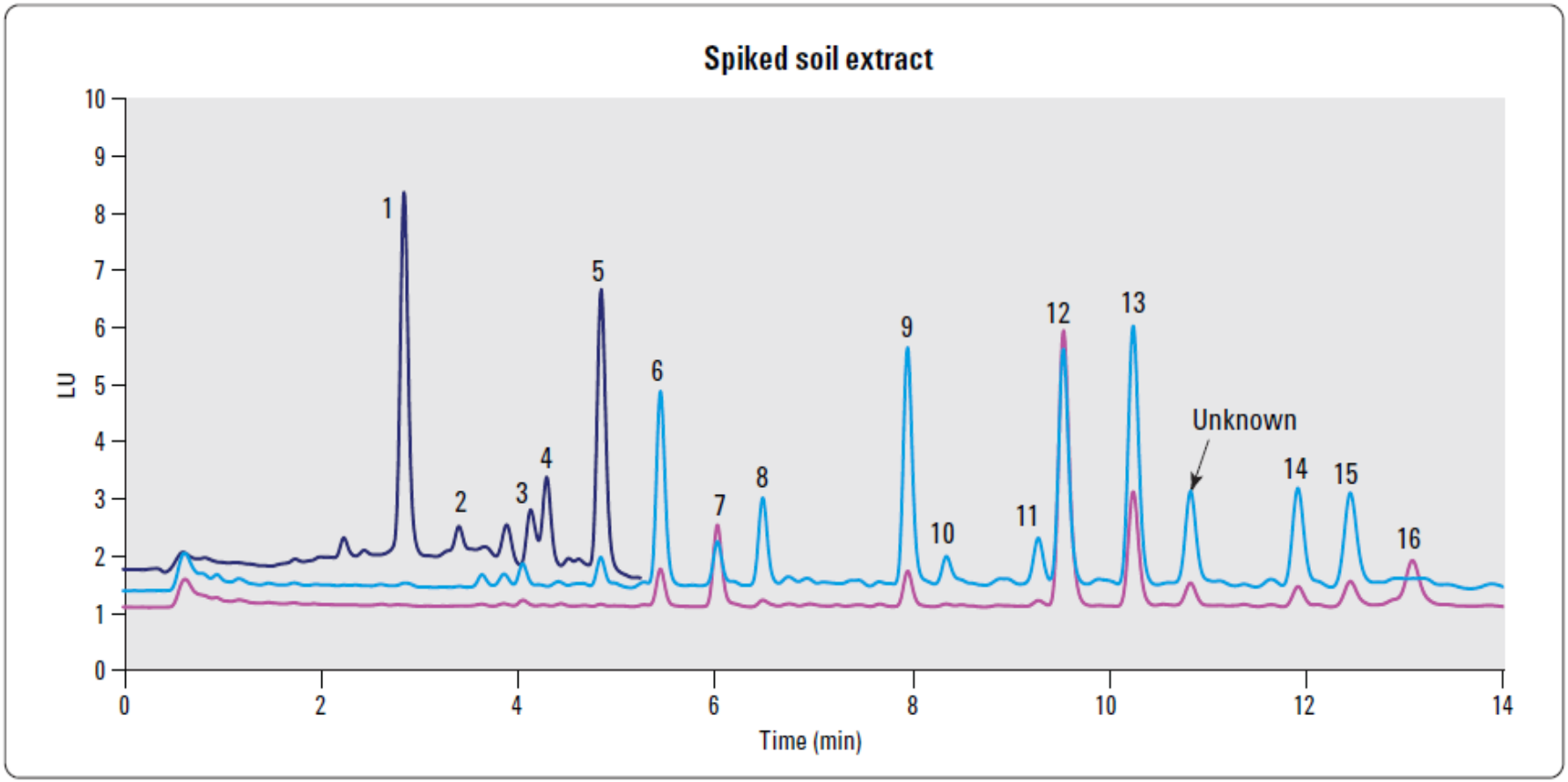
QuEChERS Extraction Procedure: Antibiotics in Liver





Recoveries for the 9 Sulfonamides in Bovine Liver

PAHs in Soil Employing QuEChERS Extraction and HPLC-FLD Analysis



Overlay HPLC – FLD chromatograms of the spiked soil sample containing: 1. Nap 2. Acy 3. Ace 4. Flu 5. Phe 6. Ant 7. Fln 8. Pyr 9. BaA 10. Chr 11. BeP 12. BeA 13. BkF 14. DahA 15. BghiP 16. InP. The spiking level for this sample was a level 1 (see Table 3). The blue portion of the chromatogram used the following excitation/emission wavelengths: 260-nm/352-nm; the red portion: 260-nm/420-nm and the light blue portion: 260-nm/440-nm.

PAHs in Soil Employing QuEChERS Extraction and HPLC-FLD Analysis: Recovery and RSDs

PAH	Spiking level (ng/g)		
	1	2	3
Naphthalene	20	100	200
*Acenaphthylene	20	100	200
Acenaphthene	10	50	100
Fluorene	10	50	100
Phenanthrene	10	50	100
Anthracene	10	50	100
Fluoranthene	10	50	100
Pyrene	10	50	100
1,2-Benzanthracene	5	20	50
Chrysene	10	50	100
Benzo[e]pyrene	5	20	50
Benz[e]acenaphthylene	5	20	50
Benzo[k]fluoranthene	5	20	50
Dibenzo[a,h]anthracene	5	20	50
Benzo[g,h,i]perylene	5	20	50
Indeno[1,2,3-cd]pyrene	5	20	50

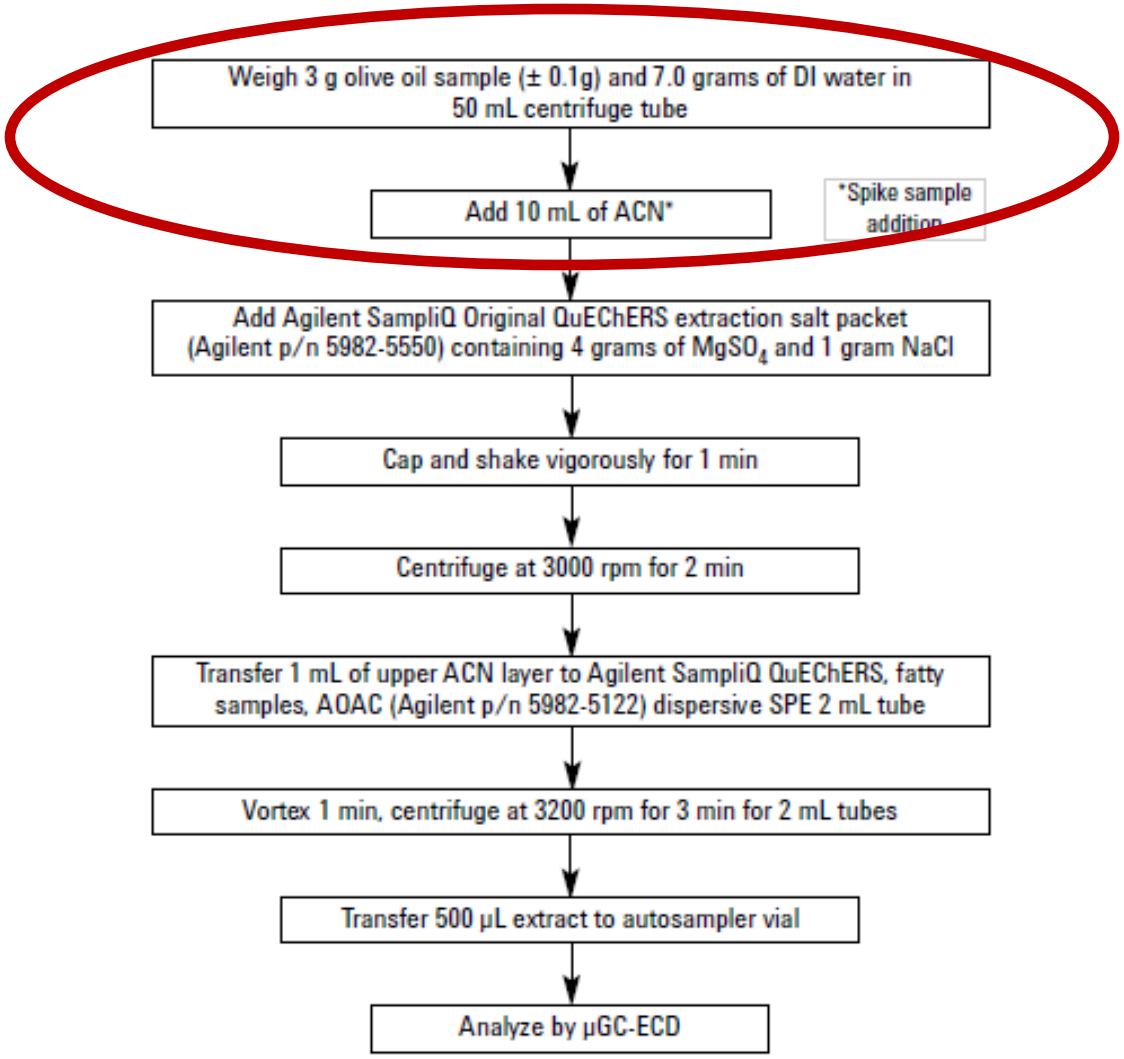
* UV detection at 230 nm

PAH	Level of spiking (ng/g) (n = 6)					
	1		2		3	
	%Recovery	%RSD	%Recovery	%RSD	%Recovery	%RSD
Naphthalene	96.5	0.7	86.2	1.4	92.8	1.4
*Acenaphthylene	87.3	0.7	90.0	1.3	91.7	1.6
Acenaphthene	91.0	1.8	89.2	1.1	89.7	1.4
Fluorene	95.2	0.8	91.4	1.3	86.0	1.2
Phenanthrene	93.0	1.0	94.6	0.7	98.1	0.9
Anthracene	91.9	1.1	90.0	0.8	97.6	0.7
Fluoranthene	93.5	1.7	94.7	1.3	87.9	1.5
Pyrene	96.3	1.3	89.4	0.9	91.2	1.9
1,2-Benzanthracene	92.9	1.7	87.8	1.5	92.8	0.7
Chrysene	98.0	1.4	92.4	1.2	95.8	1.0
Benzo[e]pyrene	97.2	1.0	97.5	0.7	90.3	0.8
Benz[e]acenaphthylene	93.2	0.9	93.1	0.6	98.0	0.7
Benzo[k]fluoranthene	94.1	1.1	97.6	0.7	91.4	1.1
Dibenzo[a,h]anthracene	89.2	1.0	99.2	1.7	90.8	1.3
Benzo[g,h,i]perylene	91.0	0.9	96.7	0.8	97.3	1.6
Indeno[1,2,3-cd]pyrene	86.0	1.2	97.8	0.8	94.3	1.3

* UV detection at 230 nm

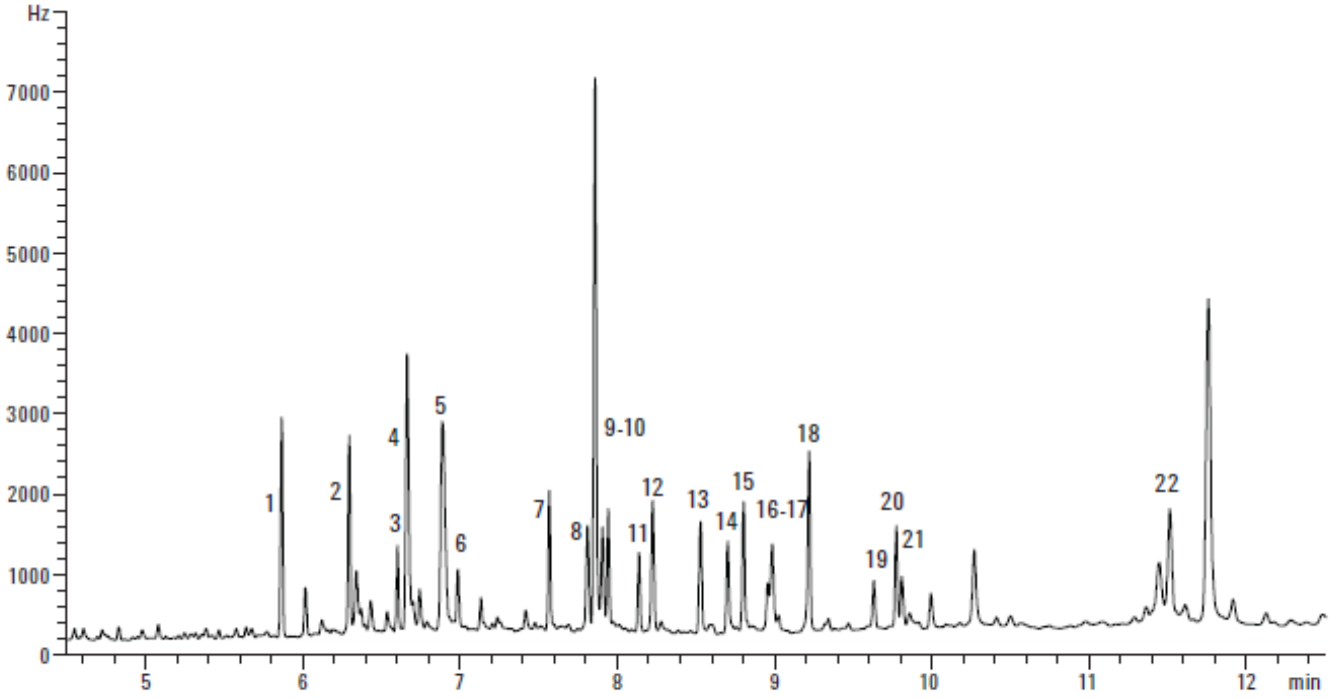
PAHs in Soil using QuEChERS extraction and Agilent 5975T GC-MS

Pesticides in Olive Oil by Dual Column GC-µECD Analysis: QuEChERS Procedure



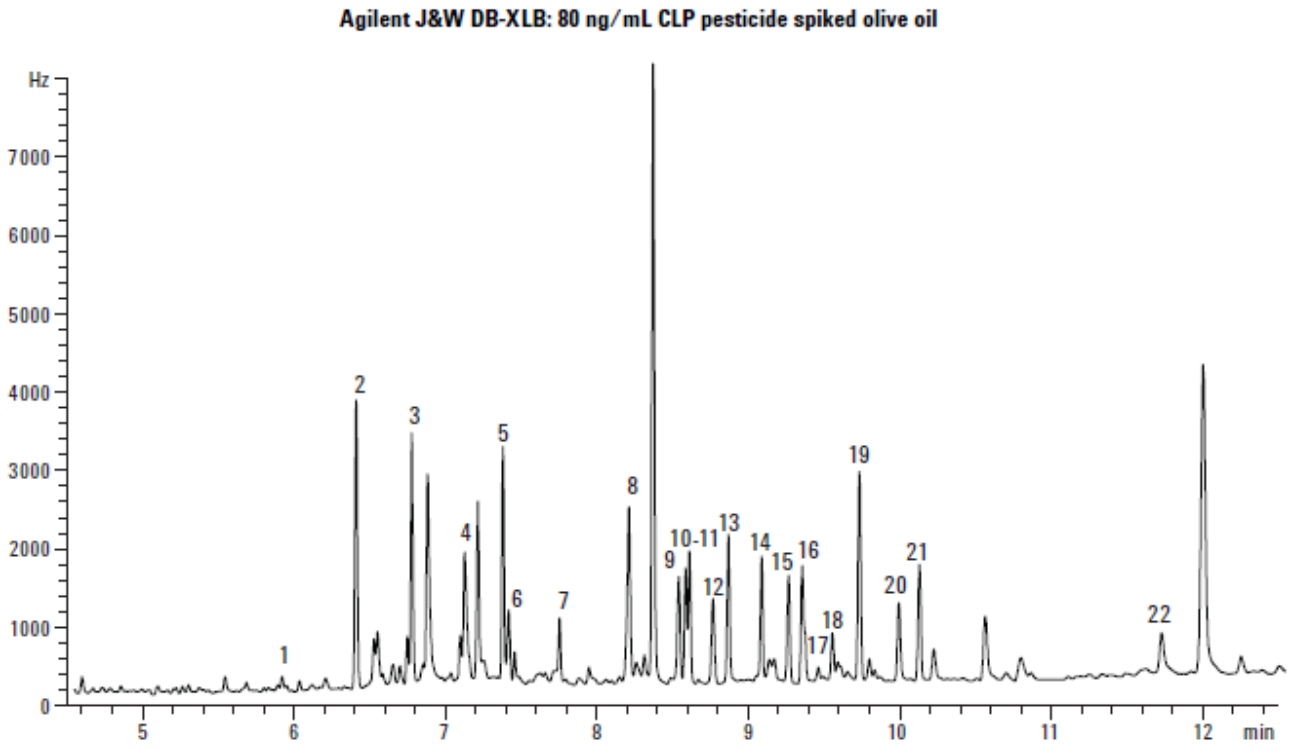
Dual Column GC-µECD Primary Column Analysis

Agilent J&W DB-35ms: 80 ng/mL CLP pesticide spiked olive oil



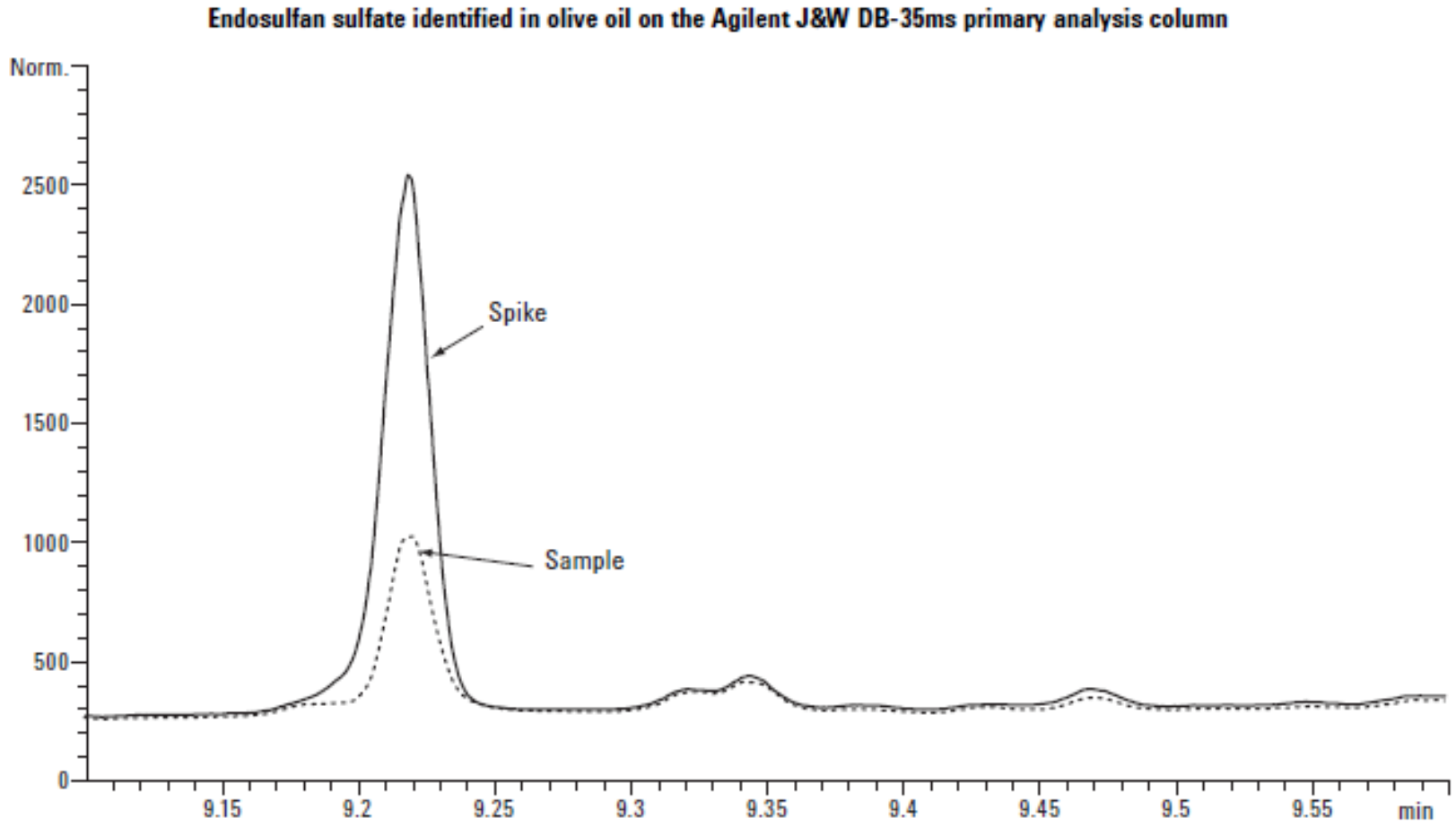
1. Tetrachloro-m-xylene (surrogate standard)
2. α-BHC
3. γ-BHC
4. β-BHC
5. Heptachlor
6. δ-BHC
7. Aldrin
8. Heptachlor epoxide
9. γ-chlordane
10. α-chlordane
11. Endosulfan I
12. Dieldrin
13. p,p-DDE
14. Endrin
15. p,p-DDD
16. Endosulfan II
17. p,p-DDT
18. Endrin aldehyde
19. Endosulfan sulfate
20. Methoxychlor
21. Endrin ketone
22. Decachlorobiphenyl (surrogate standard)

GC-μECD Second Column: Confirmation Analysis



1. Tetrachloro-m-xylene (surrogate standard)
2. α-BHC
3. γ-BHC
4. β-BHC
5. Heptachlor
6. δ-BHC
7. Aldrin
8. Heptachlor epoxide
9. γ-chlordane
10. α-chlordane
11. Endosulfan I
12. p,p-DDE
13. Dieldrin
14. Endrin
15. p,p-DDD
16. Endosulfan II
17. Endrin aldehyde
18. p,p-DDT
19. Endosulfan sulfate
20. Methoxychlor
21. Endrin ketone
22. Decachlorobiphenyl (surrogate standard)

GC-μECD Identification of Pesticide in Olive Oil



QuEChERS Extraction for Pesticides in Coffee

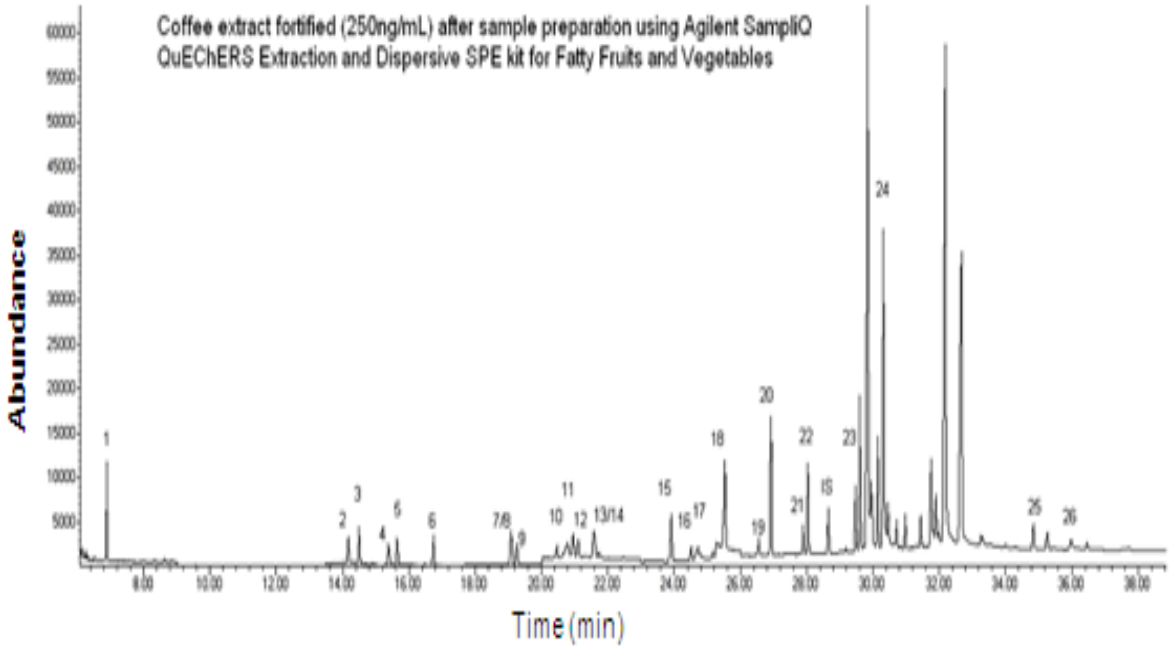
COFFEE

Figure 1: Extraction

- 10g homogenous green coffee to 10mL centrifuge tube
- Add 50µL Internal Standard (IPP 20µg/mL)
- Add10mL Hexane
- Vortex 30 seconds
- Add 10mL HPLC grade H_2O and 10mL acetonitrile
- Add 4g $MgSO_4$, 1g NaCl and two ceramic homogenizers
- Shake vigorously for 1 minute
- Centrifuge at 5000RPM for 5 minutes

Figure 2: Dispersive SPE

- Hexane layer removed and discarded
- 1mL acetonitrile layer added to 2 mL centrifuge tube
- 50mg PSA, 150mg $MgSO_4$, and 25mg C18 Added to tube
- Vortex 30 seconds
- Centrifuge 5 minutes
- 0.5mL to sampler vial
- 1.5µL injected in GCMS



Dr. Karyn Usher, West Chester University

PAHs in Fish Employing QuEChERS Extraction and HPLC-DAD Analysis: Recovery and RSDs

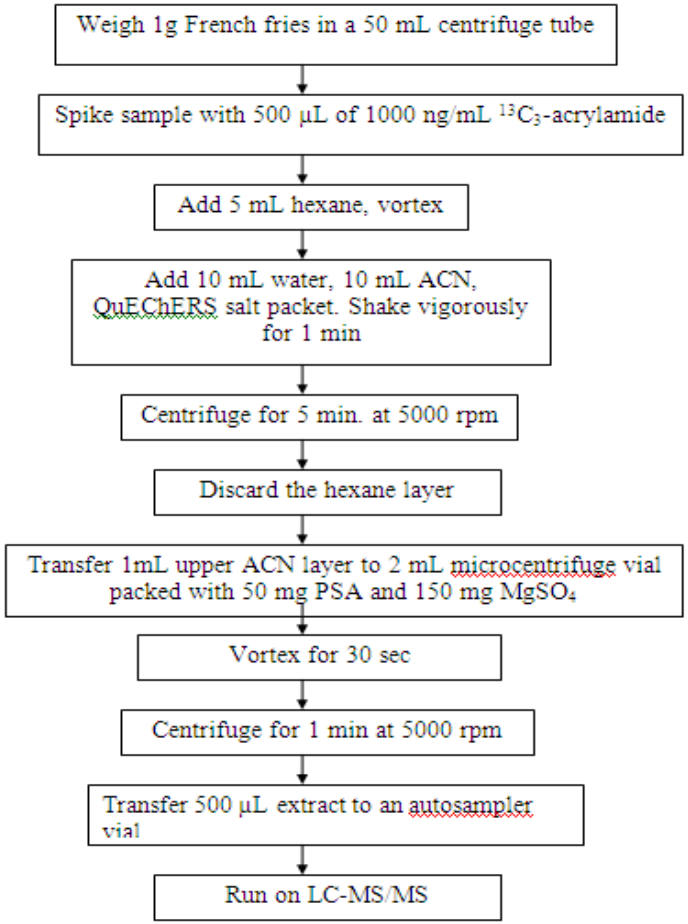
PAH	Spiking level (ng/g)		
	1	2	3
Naphthalene	20	100	200
*Acenaphthylene	20	100	200
Acenaphthene	10	50	100
Fluorene	10	50	100
Phenanthrene	10	50	100
Anthracene	10	50	100
Fluoranthene	10	50	100
Pyrene	10	50	100
1,2-Benzanthracene	5	20	50
Chrysene	10	50	100
Benzo[e]pyrene	5	20	50
Benz[e]acenaphthylene	5	20	50
Benzo[k]fluoranthene	5	20	50
Dibenzo[a,h]anthracene	5	20	50
Benzo[g,h,i]perylene	5	20	50
Indeno[1,2,3-cd]pyrene	5	20	50

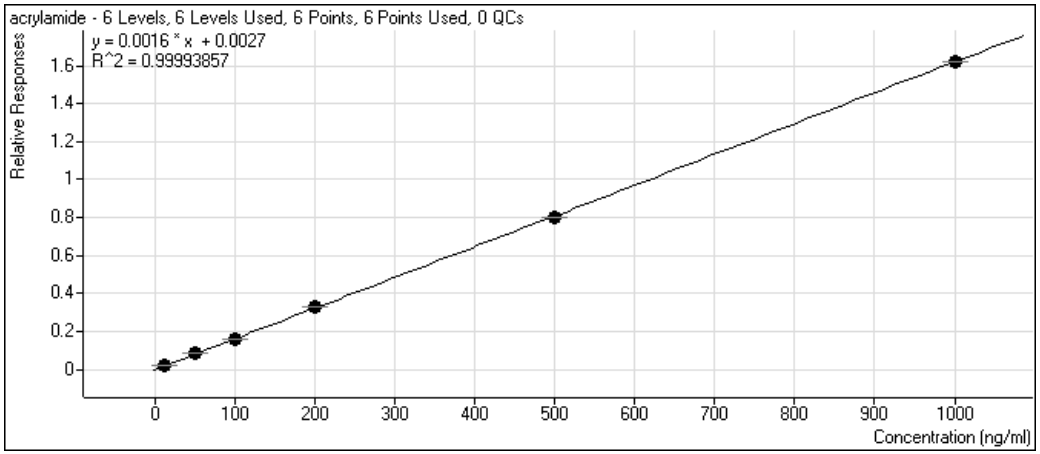
* UV detection at 230 nm

PAH	Level of spiking (ng/g) (n = 6)					
	1		2		3	
	%Recovery	%RSD	%Recovery	%RSD	%Recovery	%RSD
Naphthalene	94.7	1.4	97.9	1.1	93.8	1.4
*Acenaphthylene	87.8	1.7	96.3	1.2	85.6	0.8
Acenaphthene	92.1	1.5	93.0	1.8	96.7	0.8
Fluorene	98.1	1.5	89.9	1.0	97.2	0.9
Phenanthrene	90.6	0.9	93.8	0.8	83.1	1.7
Anthracene	96.7	1.0	87.6	0.8	92.1	0.6
Fluoranthene	83.4	1.3	93.9	1.5	95.9	1.2
Pyrene	93.5	1.8	86.1	1.3	95.0	1.4
1,2-Benzanthracene	94.5	1.3	89.6	1.6	94.9	1.0
Chrysene	101.0	1.4	97.8	1.7	87.2	1.6
Benzo[e]pyrene	88.8	1.5	85.2	1.9	95.0	1.4
Benz[e]acenaphthylene	95.5	0.7	92.7	0.7	89.2	0.9
Benzo[k]fluoranthene	93.5	0.8	94.6	0.9	98.9	0.8
Dibenzo[a,h]anthracene	88.2	0.9	97.3	1.1	97.1	0.6
Benzo[g,h,i]perylene	98.4	0.8	95.5	1.6	98.2	0.7
Indeno[1,2,3-cd]pyrene	91.5	1.5	97.9	0.9	94.3	0.7

* UV detection at 230 nm

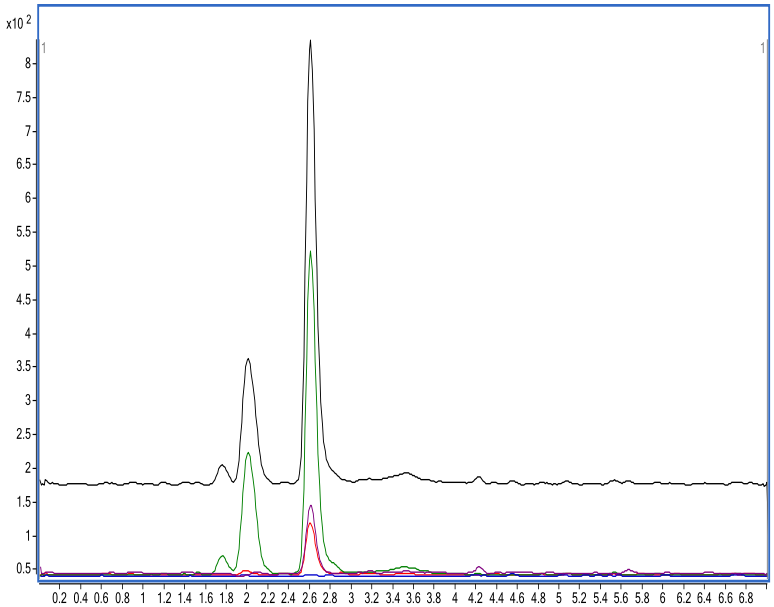
Acrylamides in Fried Foods by QuEChERS Extraction for Acrylamides





Fadwa Al-Taher, NCFST, IL

Analysis of Acrylamides in Fried Foods by QuEChERS Extraction for Acrylamides



	MRM transitions:
--------	TIC
---------	12C-AA 72.0 → 55.2
---------	12C-AA 72.0 → 26.7
---------	13C-AA 75.0 → 58.2
----------	13C-AA 75.0 → 29.0

Matrix	Concentration of acrylamide spike (ng/mL)	%Recovery (n=3)	%RSD (n=3)
1:1 ACN-water	50	116.6	4.07
1:1 ACN-water	100	114.06	4.85
French fries	100	97.14 (after blank correction)	5.04
French fries	200	97.50 (after blank correction)	2.55