

Application Guide

# FOOD TESTING SAMPLE PREPARATION

IMPROVE | CLEAN | SAVE



 **phenomenex**<sup>®</sup>  
...breaking with tradition<sup>SM</sup>

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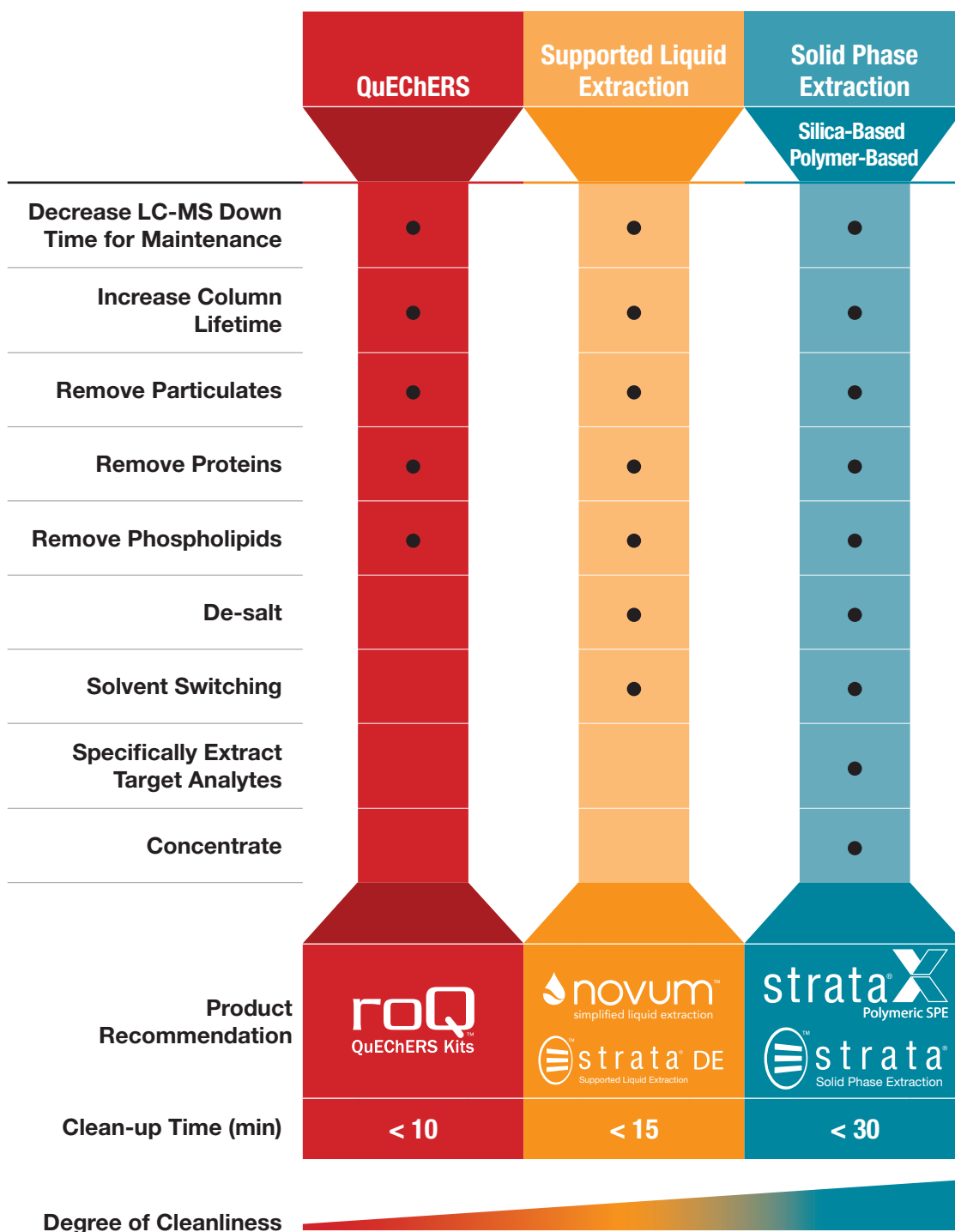
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guarantee

If Phenomenex products in this brochure do not provide at least an equivalent separation as compared to other products of the same phase and dimensions, return the product with comparative data within 45 days for a FULL REFUND.

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# Select the Appropriate Sample Prep Technique for Your Key Requirements



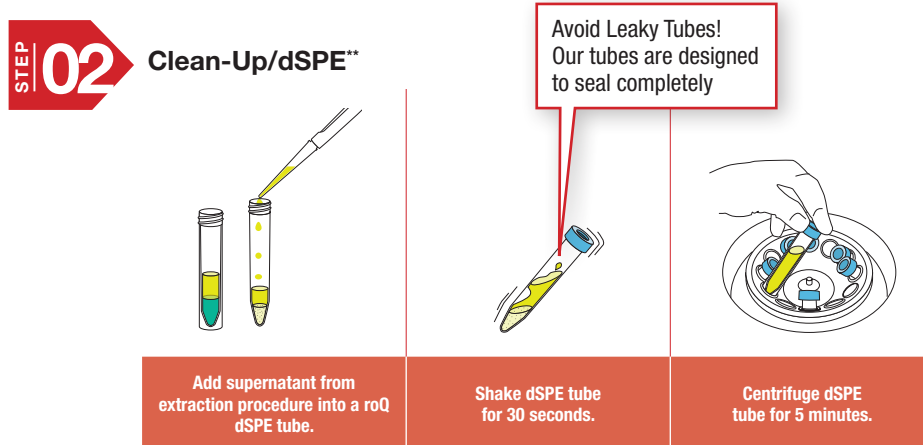
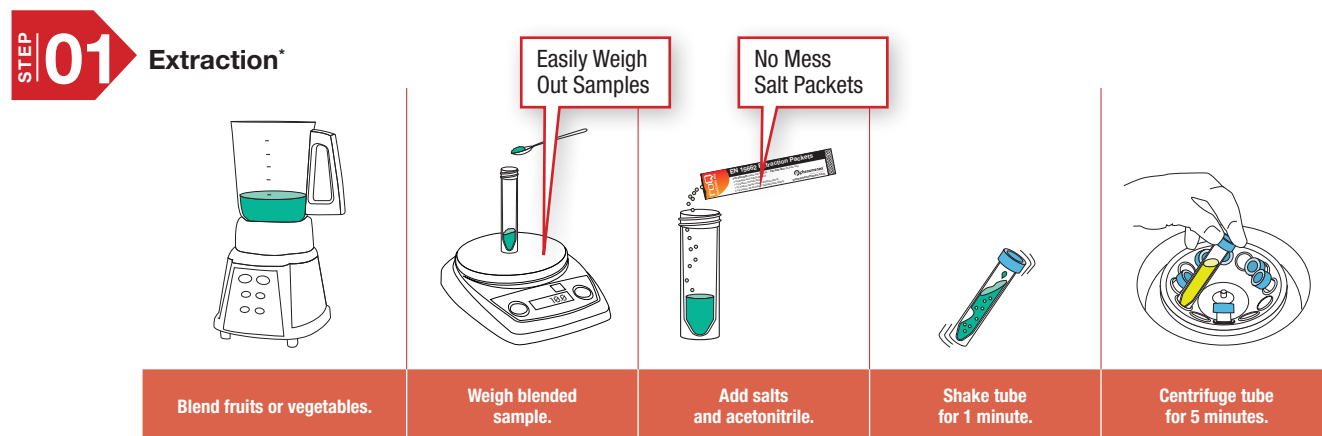
[Learn More >>](#)

# What is QuEChERS

## A Sample Preparation Technique:

- For complex sample matrices
- For analyzing a wide range of compounds
- That is **Q**uick **E**asy **C**heap **E**ffective **R**ugged and **S**afe

### How Does roQ QuEChERS Work?



\*All roQ extraction kits contain fifty easy-pour salt packets and fifty 50 mL stand-alone centrifuge tubes.

\*\*All roQ dSPE kits contain pre-weighed sorbents/salts inside 2 mL or 15 mL centrifuge tubes.



# How Does it Help Me?

QuEChERS is a simplified 2 step sample preparation process that:

- Significantly reduces sample prep time.
- Is simple, inexpensive, easy to use, effective, and rugged at extracting and cleaning samples for a wide range of compounds.

## Choose Your roQ QuEChERS Kit

### STEP 01 Extraction\*

AOAC
<b>AOAC 2007.01 Method</b> 6.0 g MgSO <sub>4</sub> , 1.5 g NaOAc <b>Part No.: KSO-8911</b>

ORIGINAL
<b>Non-Buffered Method</b> 4.0 g MgSO <sub>4</sub> , 1.0 g NaCl <b>Part No.: KSO-8910</b> 6.0 g MgSO <sub>4</sub> , 1.5 g NaCl <b>Part No.: KSO-8912</b>

EN
<b>EN 15662 Method</b> 4.0 g MgSO <sub>4</sub> , 1.0 g NaCl, 1.0 g SCTD, 0.5 g SCDS <b>Part No.: KSO-8909</b>

### STEP 02 Clean-Up/dSPE\*\*

	AOAC 2007.01		EN 15662	
	1 mL	8 mL	1 mL	6 mL
<b>General</b> 	150 mg MgSO <sub>4</sub> 50 mg PSA <b>Part No.: KSO-8920</b>	1200 mg MgSO <sub>4</sub> 400 mg PSA <b>Part No.: KSO-8928</b>	150 mg MgSO <sub>4</sub> 25 mg PSA <b>Part No.: KSO-8916</b>	900 mg MgSO <sub>4</sub> 150 mg PSA <b>Part No.: KSO-8924</b>
<b>Fats and Waxes</b> 	150 mg MgSO <sub>4</sub> 50 mg PSA 50 mg C18E <b>Part No.: KSO-8918</b>	1200 mg MgSO <sub>4</sub> 400 mg PSA 400 mg C18E <b>Part No.: KSO-8926</b>	150 mg MgSO <sub>4</sub> 25 mg PSA 25 mg C18E <b>Part No.: KSO-8913</b>	900 mg MgSO <sub>4</sub> 150 mg PSA 150 mg C18E <b>Part No.: KSO-8921</b>
<b>Pigmented</b> 	150 mg MgSO <sub>4</sub> 50 mg PSA 50 mg GCB <b>Part No.: KSO-8919</b>	1200 mg MgSO <sub>4</sub> 400 mg PSA 400 mg GCB <b>Part No.: KSO-8927</b>	150 mg MgSO <sub>4</sub> 25 mg PSA 2.5 mg GCB <b>Part No.: KSO-8914</b>	900 mg MgSO <sub>4</sub> 150 mg PSA 15 mg GCB <b>Part No.: KSO-8922</b>
<b>Highly Pigmented</b> 	—	—	150 mg MgSO <sub>4</sub> 25 mg PSA 7.5 mg GCB <b>Part No.: KSO-8915</b>	900 mg MgSO <sub>4</sub> 150 mg PSA 45 mg GCB <b>Part No.: KSO-8923</b>
<b>Pigments and Fats</b> 	150 mg MgSO <sub>4</sub> 50 mg PSA 50 mg GCB 50 mg C18E <b>Part No.: KSO-8917</b>	1200 mg MgSO <sub>4</sub> 400 mg PSA 400 mg GCB 400 mg C18E <b>Part No.: KSO-8925</b>	—	—

\*All roQ extraction kits contain fifty easy-pour salt packets and fifty 50 mL stand-alone centrifuge tubes.  
 \*\*All roQ dSPE kits contain pre-weighed sorbents/salts inside 2 mL or 15 mL centrifuge tubes.



# Multiresidue Pesticide Analysis

## Using roQ QuEChERS and LC-MS/MS

The use of a QuEChERS technique followed by LC-MS has proven to be an effective approach for screening multiresidue pesticides from various produce items including kale and grapes. Using roQ QuEChERS extraction and clean-up dSPE kits, along with the broad polarity of a Kinetex® 5 µm Biphenyl core-shell HPLC column, over 200 pesticides were screened by LC-MS/MS in under 16 minutes at concentration ranges between 0.01 ppm to 1 ppm, with the majority of analytes having a recovery range of 70-130 %.

### Easily Prepare Samples with roQ QuEChERS Kits



## Extraction

### EN 15662 Method

4.0 g MgSO<sub>4</sub>, 1.0 g NaCl,  
1.0 g SCTD, 0.5 g SCDS  
Part No.: KS0-8909

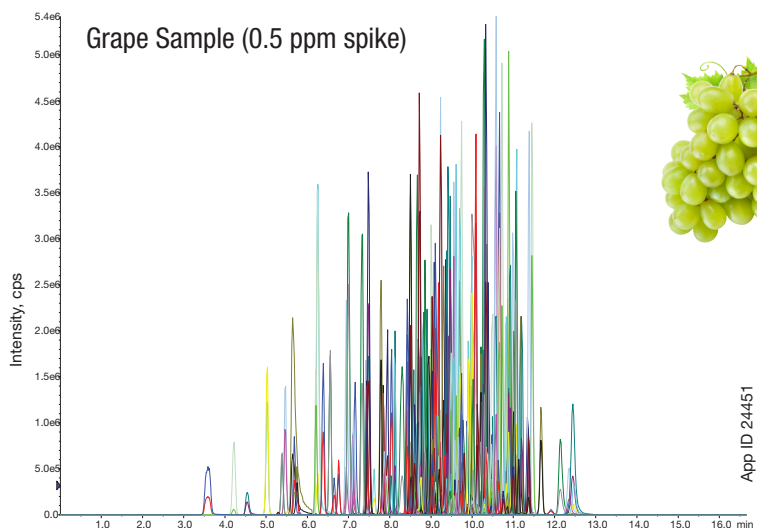
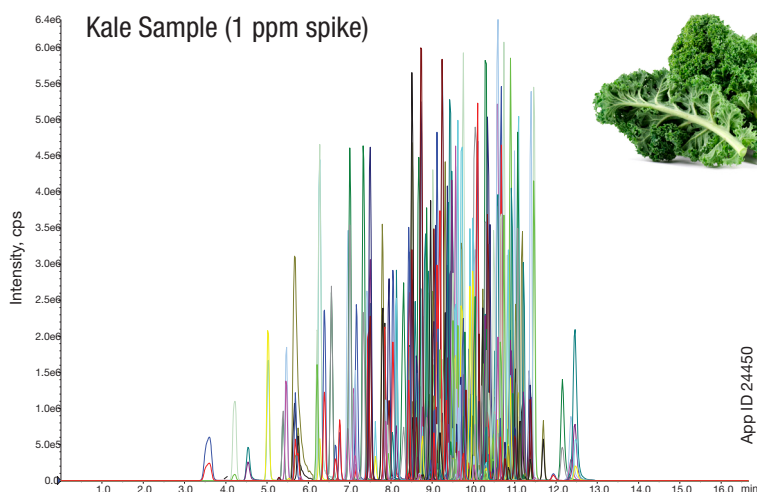


## Clean-up/dSPE

### EN 15662 Method

15 mL dSPE Kit  
900 mg MgSO<sub>4</sub>, 150 mg PSA, 15 mg GCB  
Part No.: KS0-8922

### Great Selectivity for Polar and Non-Polar Pesticides



### LC-MS/MS Conditions

<b>Column:</b>	Kinetex 5 µm Biphenyl 100 Å	
<b>Dimensions:</b>	50 x 4.6 mm	
<b>Part No.:</b>	00B-4627-E0	
<b>Guard:</b>	SecurityGuard™ ULTRA Cartridges: AJ0-9207	
<b>Mobile Phase:</b>	A: 10 mM Ammonium formate in Water B: Methanol	
<b>Gradient:</b>	<b>Time (min)</b>	<b>% B</b>
	0	2
	1	2
	10	100
	13	100
	13.1	2
	16	2
<b>Injection:</b>	20 µL	
<b>Flow Rate:</b>	0.5 mL/min	
<b>Temperature:</b>	35 °C	
<b>Detection:</b>	MS/MS (SCIEX 4000 QTRAP®)	
<b>Sample:</b>	Pesticide Mix	

To learn more about roQ QuEChERS, visit

[www.phenomenex.com/roQ](http://www.phenomenex.com/roQ)

# Mycotoxins from Cereal Products

## Using roQ QuEChERS and LC-MS/MS

Exposure to mycotoxins through consumption, inhalation, or dermal routes can result in a variety of health effects. Different types of mycotoxins, depending on the intended use, have acceptable levels allowable by the FDA. Due to the complexity, a specific and sensitive method is necessary to proactively monitor mycotoxins in food. Demonstrated is a rapid sample preparation and LC-MS/MS method for screening all major mycotoxins from corn-meal using roQ QuEChERS kits and a Kinetex® 2.6 µm XB-C18 core-shell HPLC column.

### Easily Prepare Samples with roQ QuEChERS Kits



### Extraction

#### EN 15662 Method

4.0 g MgSO<sub>4</sub>, 1.0 g NaCl,  
1.0 g SCTD, 0.5 g SCDS  
Part No.: KS0-8909



### Clean-up/dSPE

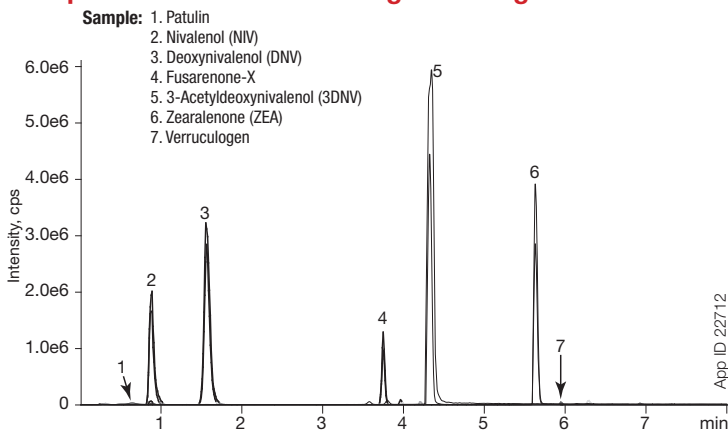
#### EN 15662 Method

15 mL dSPE Kit  
900 mg MgSO<sub>4</sub>, 150 mg PSA  
Part No.: KS0-8924

### Reconstitution

2 mL of supernatant from the dSPE step was evaporated to dryness under a stream of Nitrogen at 60°C. The sample was reconstituted in 0.25 mL of 0.5% Acetic acid/Methanol (95:5) for analysis.

### Sample Extract Ion Chromatogram - Negative Mode



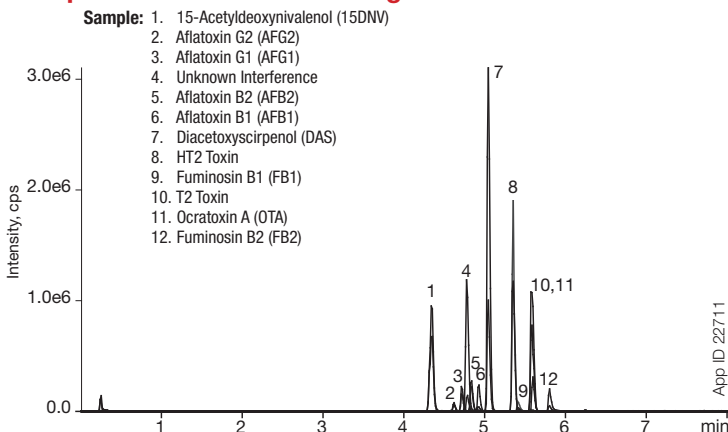
### LC-MS/MS Conditions

**Column:** Kinetex 2.6 µm XB-C18  
**Dimensions:** 50 x 2.1 mm  
**Part No.:** 00B-4496-AN  
**Guard:** SecurityGuard™ ULTRA Cartridge: AJ0-8782  
**Mobile Phase:** A: 5 mM Ammonium acetate with 0.5% Acetic acid  
B: 5 mM Ammonium acetate in Methanol with 0.5% Acetic acid

Gradient:	Time (min)	% B
	0	5
	2	5
	5	80
	5.2	98
	8	98

**Injection:** 10 µL  
**Flow Rate:** 0.45 mL/min  
**Temperature:** 22°C  
**Detection:** MS/MS (SCIEX API 5000™)

### Sample Extract Ion Chromatogram - Positive Mode



### Recovery of Mycotoxins

Analyte	% Recovery	Analyte	% Recovery
AFB1	73	FB1	8
AFB2	87	FB2	14
AFG1	67	Patulin	118
AFG2	93	ZEA	91
HT2 Toxin	96	DNV	80
T2 Toxin	91	NIV	66
DAS	90	3DNV	96
OTA	79	Fusarenone-X	83
15DNV	99	Verruculogen	81

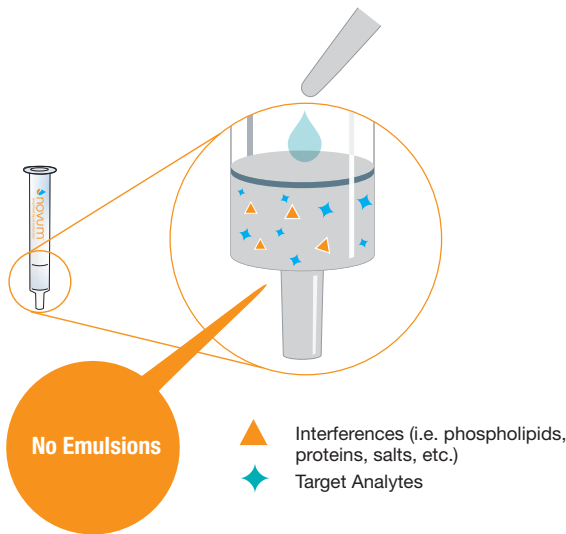
# Supported Liquid Extraction

**Supported Liquid Extraction (SLE) is a faster, easier, and more reliable way to perform liquid-liquid extractions**

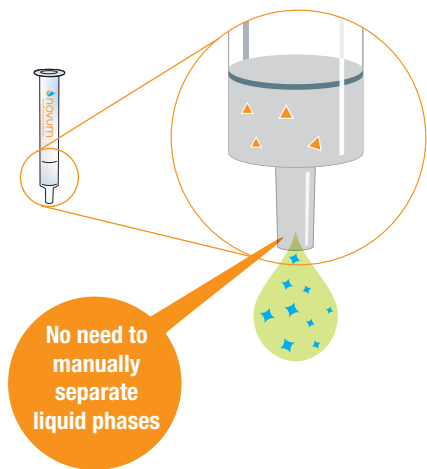
- Eliminates interferences from your analysis, such as proteins and phospholipids, without performing extensive method development
- Novum synthetic SLE provides consistent, reliable results from lot-to-lot
- Strata DE diatomaceous earth SLE is an alternative to other diatomaceous earth SLE products available

## An Easy, Automatable Procedure

**STEP 01** Load Your Sample in Aqueous Solvent



**STEP 02** Collect Your Target Analytes in Water Immiscible Solvent



## Determine Which SLE Sorbent is Right for Your Extraction



<b>Synthetic</b>	<b>Sorbent Type</b>	<b>Diatomaceous Earth</b>
<b>Lot-to-lot consistency and reproducibility</b>	<b>Advantages</b>	<b>Cost effective and large volume capabilities</b>
<b>Ethyl Acetate, MTBE</b>	<b>Extraction Solvents</b>	<b>Hexane, DCM, MTBE Ethyl Acetate</b>
<b>MINI 96-Well Plates, MAX 96-Well Plates</b>	<b>Plate Formats</b>	<b>200 µL 96-Well Plates, 400 µL 96-Well Plates</b>
<b>1 cc, 3 cc, 6 cc, 12 cc</b>	<b>Tube Formats</b>	<b>12cc and 60 cc</b>

SLE sorbent selections are dependent on extraction solvents and sample volume. Contact your Phenomenex representative to learn which SLE product is right for you!



# Acrylamide from Coffee

## Using Novum SLE

Acrylamide is commonly found in foods that are made from plants such as potato products, grain products, and coffee whose preparation often requires longer cooking times and higher temperatures. Below we explore how to use Novum SLE tubes to clean up a coffee matrix in order to quantitate known acrylamide levels.

### Sample Pre-treatment

Prepared control coffee was left on the bench to reach room temperature before further pre-treatment.

Ground Coffee Control (40mg/mL)

- 60g of ground coffee was percolated with 1500mL of boiling water
- Instant Coffee Control (8mg/mL)
- 2g of instant coffee was dissolved in 250mL of boiling water

Acrylamide standard was spiked into control coffee to reach 100ng/mL (ground coffee) and 200ng/mL (instant coffee) by adding 20µL Acrylamide-13C3 (4µg/mL in water) to 800µL of the prepared coffee.

### SLE Protocol

<b>Cartridge:</b>	Novum 6 cc Tube
<b>Part No.:</b>	8B-S138-JCH
<b>Add:</b>	150 µL 2% Ammonium hydroxide in Water to the spiked samples (from pre-treatment step), vortex for 30 seconds.
<b>Load:</b>	Pre-treated sample onto the Novum 6 cc tubes and apply a short and gentle pulse of vacuum (~5-10 seconds at 5" or less of Hg) until the sample has completely entered the media. Wait for 5-6 minutes. Note: Inadequate or excessive wait periods can lead to variable recoveries and poor precision.
<b>Elute:</b>	2x 2.5 mL Ethyl acetate/Tetrahydrofuran (1:1) and collect under gravity into a collection tube that contains 10 µL Ethylene glycol. Apply vacuum at 5" of Hg (or lower) for 20-30 seconds to complete extraction. Note: To reduce analyte loss due to dry down, ethylene glycol was added to the collection tube to prevent the sample from drying completely during the dry down step.
<b>Dry Down:</b>	Evaporate samples to complete dryness under a slow stream of Nitrogen at 45 °C.
<b>Reconstitute:</b>	300 µL Water

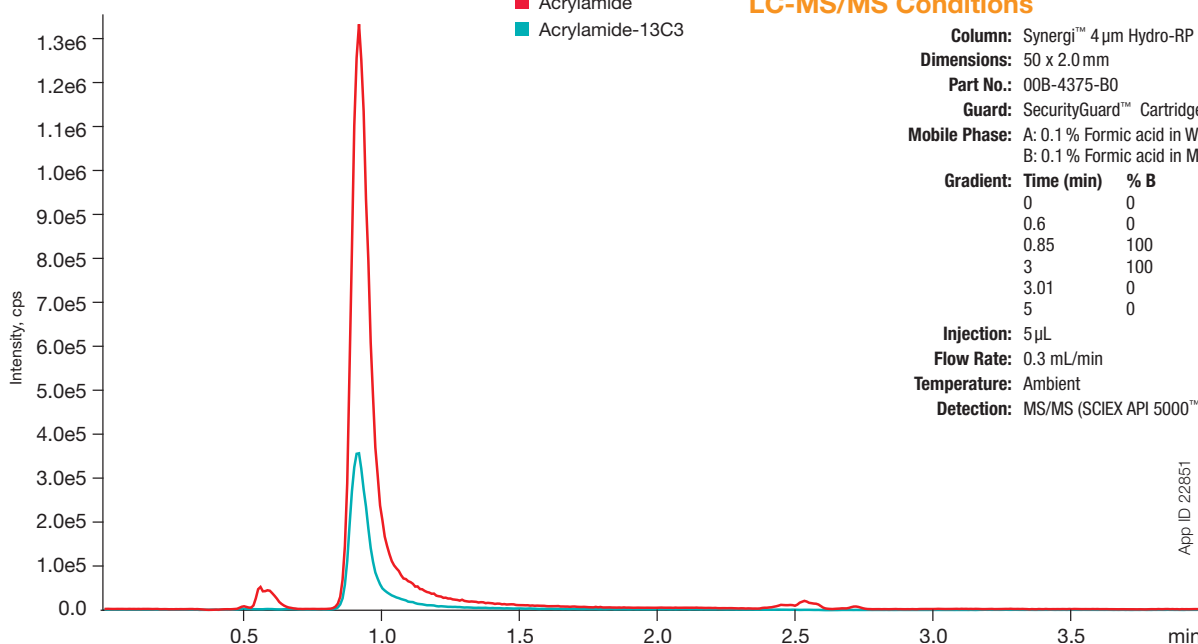
### Recovery of Acrylamide

Sample ID	Ground coffee (100 ng/mL)	Instant coffee (200 ng/mL)
Mean of area ratio	1.89	3.75
STDV	0.01	0.06
CV (%)	0.78	1.61
Absolute Recovery (%)	94.9	92.8
n=	6	6

High recoveries and precision using Novum SLE!

### Acrylamide from Coffee (100ng/mL)

■ Acrylamide  
■ Acrylamide-13C3



### LC-MS/MS Conditions

**Column:** Synergi™ 4 µm Hydro-RP  
**Dimensions:** 50 x 2.0 mm  
**Part No.:** 00B-4375-B0  
**Guard:** SecurityGuard™ Cartridge: AJ0-7510  
**Mobile Phase:** A: 0.1% Formic acid in Water  
 B: 0.1% Formic acid in Methanol  
**Gradient:**

Time (min)	% B
0	0
0.6	0
0.85	100
3	100
3.01	0
5	0

**Injection:** 5 µL  
**Flow Rate:** 0.3 mL/min  
**Temperature:** Ambient  
**Detection:** MS/MS (SCIEX API 5000™), ESI+

App ID 22851

# Sterols in Olive Oil

## Using Strata DE SLE, SPE and GC-FID

Due to frequent adulteration, a reliable and efficient method was developed to determine the concentration of sterols in olive oil, which can confirm the classification of olive oil. Presented is a modified International Olive Council (IOC) method for sterol determination which was improved upon using diatomaceous earth SLE (Strata DE) followed by further clean up by Solid Phase Extraction (SPE) to remove hydrocarbons as well as more polar interferences from the solution. The isolated sterols and triterpene alcohols are then derivatized as the trimethylsilyl ethers prior to GC-FID analysis. The result is an improved method for determining sterols, erythrodiol and uvaol in olive oil.

### Internal Standard Preparation

Add 40 µL of 1 mg/mL cholestanol in chloroform to a clean, dry 20 mL screw-top test tube and evaporate to dryness under a slow stream of Nitrogen.

### Saponification

1. Add 200 mg of olive oil sample to the test tube containing the internal standard.
2. Add 1.5 mL of 2M Potassium hydroxide in 95% Ethanol.
3. Cap the tube and heat in an 80 °C oven for 25 minutes.
4. Mix sample gently to ensure homogeneity (sample should appear as a clear solution) and continue heating for an additional 25 minutes.
5. After heating, add 13.5 mL of deionized water and mix. The entire diluted volume is now ready to load onto the SLE cartridge.

### SLE Protocol

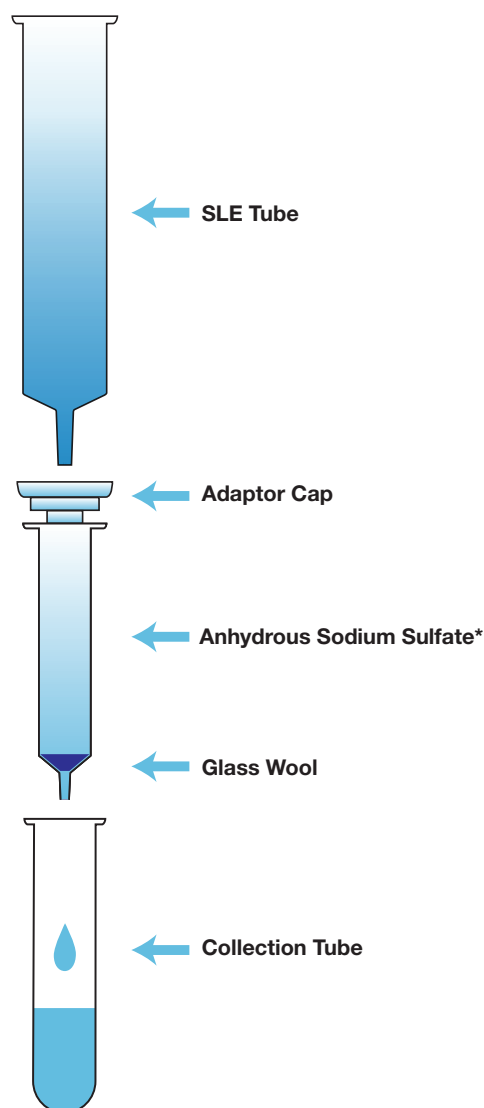
<b>Cartridge:</b>	Strata DE SLE Tube, 60 cc Tube (20 mL loading capacity)
<b>Part No.:</b>	8B-S325-VFF
<b>Load:</b>	Diluted sample (from saponification step 5) plus 2x 1 mL DI water rinse (17 mL total volume, gravity flow)
<b>Wait:</b>	15 minutes
<b>Extract:</b>	3x 15 mL Diethyl ether (gravity flow)
<b>Evaporate:</b>	Dry under Nitrogen at 40 °C (greenish-yellow, oily residue)
<b>Reconstitute:</b>	5 mL of Hexane

### SPE Protocol and Derivatization

<b>Cartridge:</b>	Strata Si-1 Tube, 1 g/6 mL
<b>Part No.:</b>	8B-S012-JCH
<b>Condition:</b>	1. 2x 6 mL Hexane 2. 1 mL 0.2M Potassium hydroxide in 95% Ethanol
<b>Equilibrate:</b>	5 mL Hexane (immediately after Potassium hydroxide elution)
<b>Load:</b>	Reconstituted SLE extract plus 2x 1 mL Hexane rinse from the SLE collection tube (7 mL total volume)
<b>Wash:</b>	85 mL Hexane/Diethyl ether (98:2) under 3" Hg vacuum, flow rate of 2 mL/min*
<b>Elute:</b>	10 mL Hexane/Diethyl ether (60:40)
<b>Dry:</b>	Dry under Nitrogen at 50 °C. After evaporating to dryness, add 3-4 drops of acetone and then re-evaporate under Nitrogen to remove any occluded water. Place in 200 °C oven for 10 minutes.
<b>Derivatization:</b>	250 µL Pyridine/BSTFA (3:1) at 80 °C for 30 minutes

\* To handle the large volume of eluant, a 60 mL empty reservoir tube was attached to the 6 mL SPE tube.

### SLE setup with sodium sulfate drying tube attached to an SLE column (Gravity Flow)



\*Sodium sulfate tubes were used as a sample drying step.

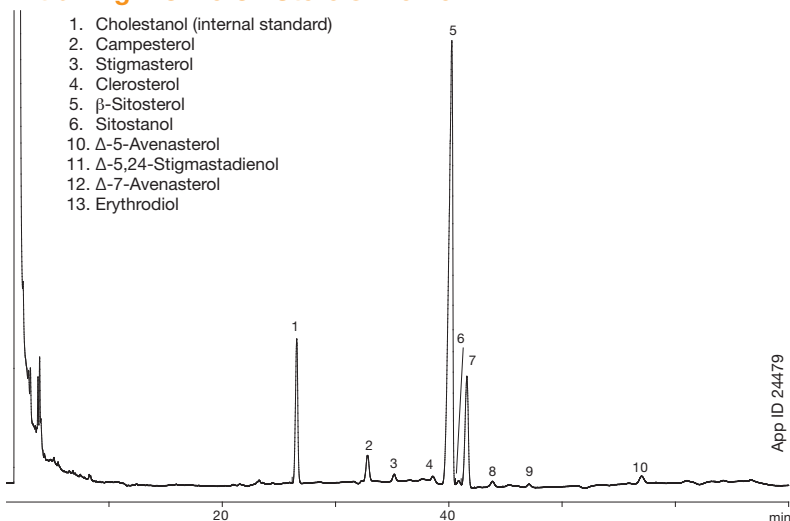
To read full technical note, go to:

[www.phenomenex.com/foodresources](http://www.phenomenex.com/foodresources)

# Sterols in Olive Oil (cont'd)

## Using Strata DE SLE, SPE, GC-FID

### Extra Virgin Olive Oil Sterols Profile



App ID 24479

### GC-FID Conditions

**Column:** Zebron™ ZB-5PLUS™

**Part No.:** 7HG-G032-11

**Dimensions:** 30 meter x 0.25 mm x 0.25 μm

**Injection:** Split 5:1 @ 280 °C, 1 μL

**Recommended Liner:** Zebron PLUS Single Taper Z-Liner™

**Liner Part No.:** AG2-0A13-05 (for Agilent® systems)

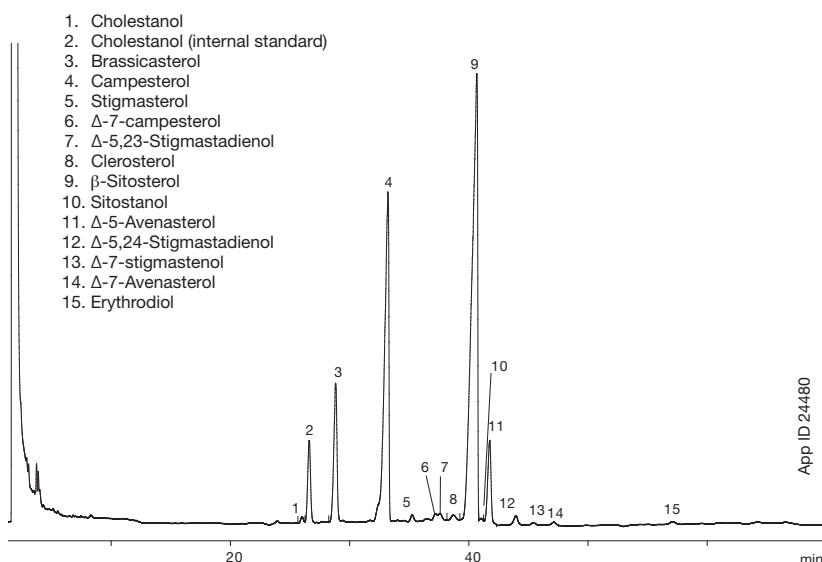
**Carrier Gas:** Helium @ 0.9 mL/min (constant flow)

**Oven Program:** 260 °C for 70 min

**Detector:** FID @ 300 °C

**Samples:** Analytes were derivatized with BSTFA in pyridine (1:3)

### Adulterated Olive Oil: Extra Virgin Olive Oil/Canola Oil (50:50)



App ID 24480

### IOC Virgin Olive Oil Sterol Criteria

Standard Name	IOC Standard Criteria for Virgin Olive Oil	Extra Virgin Olive Oil		Adulterated Olive Oil	
		% Recovery	% RSD (n=3)	% Recovery	% RSD (n=2)
Apparent β-Sitosterol*	≥ 93.0 % of total sterols	94.6 %	0.3	60.3 %	1.2
Cholesterol	≤ 0.5 % of total sterols	not detected	-	0.3 %	13.3
Brassicasterol	≤ 0.1 % of total sterols	not detected	-	8.7 %	0.7
Campesterol	≤ 4.0 % of total sterols	3.8 %	6.8	29.1 %	1.3
Stigmasterol	≤ Campesterol (≤ 4.0 % of total sterols)	1.0 %	9.0	0.6 %	45.0
Δ-7-Stigmastenol	≤ 0.5 % of total sterols	not detected	-	0.7 %	2.9
Uvaol + Erythrodiol	≤ 4.5 % of total sterols	1.8 %	31	0.3 %	1.0
<b>Total Sterols</b>	<b>≥ 1000 mg/kg</b>	<b>1324 mg/kg</b>	<b>6</b>	<b>4221 mg/kg</b>	<b>1.0</b>

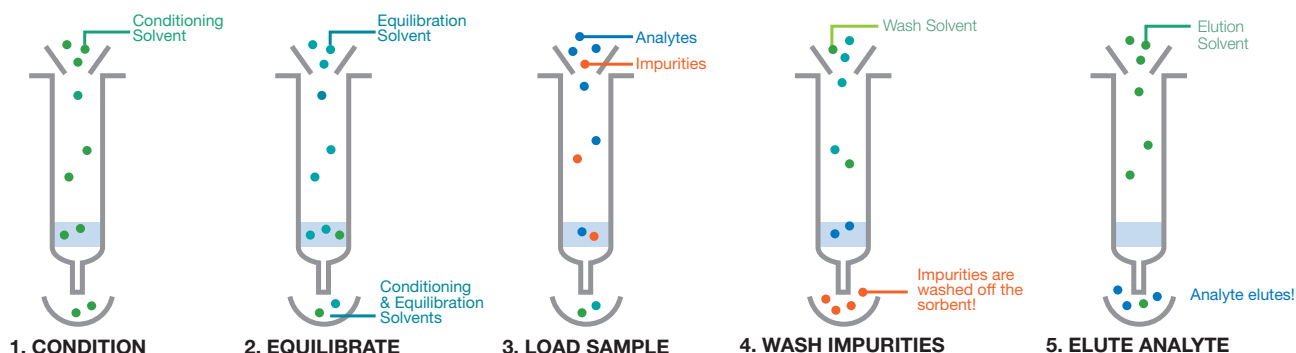
\*Apparent β-sitosterol = β-sitosterol + Δ-5-avenasterol + Δ-5,23-stigmastadienol + clerosterol + sitostanol + Δ-5,24-stigmastadienol. Total sterols = cholesterol + 24-methylene cholesterol + brassicasterol + campesterol + campestanol + stigmasterol + Δ-7-campesterol + Δ-5,23-stigmastadienol + apparent β-sitosterol + Δ-7-avenasterol.

# Solid Phase Extraction (SPE)

SPE is a very targeted form of sample preparation that allows you to isolate your analyte of interest while removing any interfering compounds that may be in your sample.


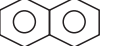

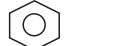
- Targeted analyte extraction for cleaner analysis
- Concentration of samples for better chromatographic results
- Solvent switching for GC or LC compatibility

## SPE General Protocol



## SPE Retention Mechanisms

Identify the general mechanism, then isolate the most specific Strata or Strata-X sorbent by matching the analyte functional groups to the sorbent functional group.

Sample Matrix		SPE Mechanism	Analyte Functional Group	Sorbent Functional Group	Strata-X Polymeric SPE Sorbent	Strata Silica-Based SPE Sorbent
Aqueous	Add H <sub>2</sub> O	Reversed Phase	R  hydrocarbon  aromatic	R  hydrocarbon  aromatic	X, XL	C18-E, C18-U, C8 C18-T PH, SDBL
Organic (Water Miscible)	For ionic species only	Ion-Exchange	NR <sub>4</sub> <sup>+</sup> strong RNH <sub>3</sub> <sup>+</sup> weak RSO <sub>3</sub> <sup>-</sup> strong RCO <sub>2</sub> <sup>-</sup> weak	-O <sub>2</sub> C- weak -O <sub>3</sub> S- strong +H <sub>3</sub> N- weak +R <sub>3</sub> N- strong	X-CW, XL-CW XL-C, X-C X-AW, XL-AW X-A, XL-A	WCX Screen-C, SCX NH <sub>2</sub> Screen-A, SAX
Organic (Water Immisible)	Add Hexane	Normal Phase	R - OH hydroxyl R - NH <sub>2</sub> amino	CN polar OH polar		CN, NH <sub>2</sub> Si-1, CN, EPH

To find the right SPE sorbent for your analysis, go to [www.phenomenex.com/SPE](http://www.phenomenex.com/SPE)

# Chlorinated Pesticides in Poultry Tissue

## Using Strata SPE and GC-ECD

Animals used for food consumption are exposed to contaminants at levels that can pose harm to the human population. Presented is a method developed using Strata Alumina-N SPE and GC-ECD for pesticides analysis from poultry fat. This method improves upon the traditional procedure by reducing time and increasing accuracy and reliability.

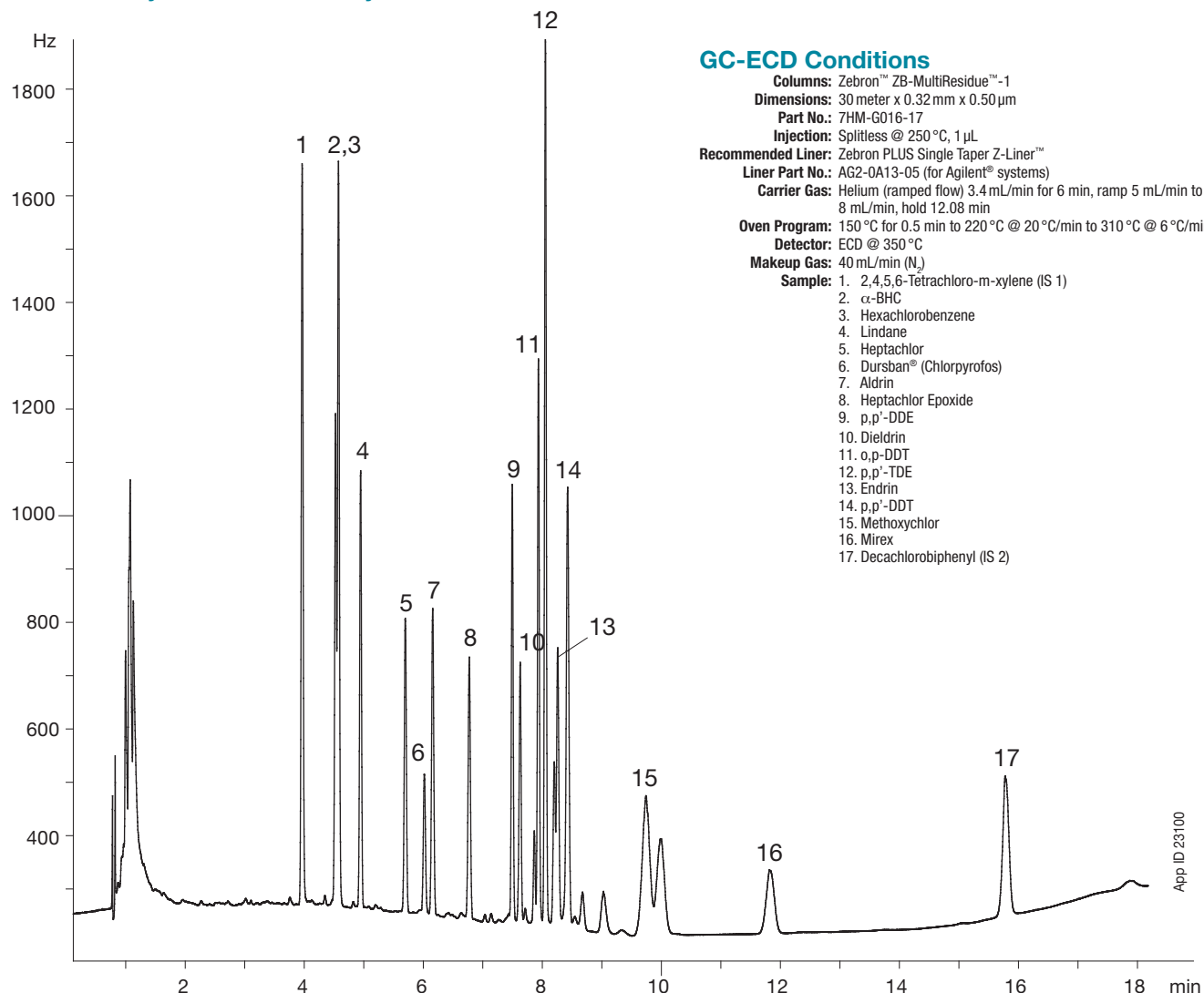
### Pre-treatment Protocol

1. Using 1 minute intervals with a microwave, render poultry fat pads ensuring the sample does not exceed 100°C
2. Weigh 1 gram of rendered fat into a 10mL volumetric flask and bring to volume with hexane containing internal standards 1 and 2 (see analyte 1 and 17)
3. Vortex or shake volumetric flasks to ensure proper mixing

### SPE Protocol

<b>Cartridge:</b>	Strata Alumina-N, 2 g/12 mL
<b>Part No.:</b>	8B-S313-KDG
<b>Condition:</b>	Methanol/Water (86:14) at 10 mL/min
<b>Equilibrate:</b>	Petroleum ether at full cartridge volume at 10 mL/min
<b>Load:</b>	1 mL Pre-treated sample
<b>Elute:</b>	Ethyl ether/Petroleum ether (1.5:98.5) at full cartridge volume and collect eluent
<b>Dry Down:</b>	Dry down at ambient temperatures under a stream of Nitrogen and evaporate to dryness
<b>Reconstitute:</b>	2 mL Hexane

### GC-ECD Analysis of Chlorinated Hydrocarbons



# Vitamin D<sub>2</sub>/D<sub>3</sub> and Pre-D<sub>2</sub>/D<sub>3</sub> in Pet Food

## Using Strata SPE and LC-MS/MS

Vitamin D is an essential fat soluble vitamin, most known for its requirement in bone formation in animals. The consensus is that Vitamin D<sub>3</sub> is the most biologically active form of Vitamin D, and thus needs to be distinguished apart from Vitamin D<sub>2</sub> for testing purposes. The challenges in the determination of Vitamin D<sub>2</sub>/D<sub>3</sub>, such as matrix interferences, isomer conversions, and light sensitivity are still persistent in pet food. Presented is an LC-MS/MS method with a simplified saponification and extraction procedure suitable for a variety of pet food samples.

### Saponification/Pre-extraction/LLE Extraction Procedure:

- 1) Weight ~2g blended dog food powder + 0.2g Ascorbic acid + 0.2g Pyrogillic acid into 1st 50mL centrifuge tube, mix, spike with corresponding standard stock solution to make STDs and QCs
- 2) Add 100µL of IS stock mix (10/10µg/mL of Vit D<sub>2</sub>-d<sub>3</sub>/Vit D<sub>3</sub>-d<sub>3</sub>) and 8mL Ethanol to all samples except blank, mix/shake for the pre-extraction at 200rpm for 15min
- 3) Add 2mL of 45% Potassium hydroxide to all samples, mix/shake/incubate at 200rpm under room temperature for 1 hour
- 4) Add 5mL DI water to 1st 50mL centrifuge tube, shake at 200 rpm for 15 min, centrifuge at 4500rpm for 5 min, transfer upper supernatant (dark brown color) to 2nd 50mL centrifuge tube
- 5) Add 5mL 40% Ethanol to 1st 50mL centrifuge tube, shake at 200rpm for 15 min, centrifuge at 4500rpm for 5 min, transfer/combine upper supernatant (dark brown color) to corresponding 2 nd 50mL centrifuge tube, discard 1st 50mL centrifuge tube
- 6) Add 6mL Heptane to 2 nd 50mL centrifuge tube, shake at 200 rpm for 15 min, centrifuge at 4500rpm for 5 min, transfer upper supernatant (light yellow color) to a 100 X 16mm glass tube by plastic transfer pipettes, dry samples in TurboVap® at 30°C under Nitrogen for 20min
- 7) Add 6mL Heptane to 2 nd 50mL centrifuge tube again, shake at 200rpm for 15 min, centrifuge at 4500rpm for 5 min, transfer upper supernatant (light yellow color) to corresponding 100 X 16mm glass tube by plastic transfer pipettes, discard 2 nd 50mL centrifuge tube, dry samples in TurboVap at 30°C under Nitrogen about 10-15 min until liquid volume remaining is ~3mL (DO NOT DRY DOWN ALL LIQUID VOLUME)

### SPE Protocol

**Cartridge:** Strata NH<sub>2</sub> (55µm, 70A), 2g/12 mL Giga™ Tubes

**Part No.:** 8B-S009-KDG

**Condition:** 2x 6 mL Methylene chloride (DCM)

**Equilibration:** 2x 6 mL Heptane/Hexane (50:50)

**Load:** ~3 mL supernatant from LLE (procedure step 7)

**Wash:** 2x 5 mL 10% DCM in Hexane

**Dry:** 4-5 min

**Elute:** 2x 6 mL Methylene chloride (DCM) into 10 X 16 mm glass tube

**Dry Down:** Under Nitrogen at 30°C

**Reconstitute:** 300 µL of Ethanol/Water (40:60)

### LC-MS/MS Conditions

**Column:** Kinetex® 2.6µm F5

**Dimensions:** 30 x 2.1 mm

**Part No.:** 00A-4723-AN

**Guard:** SecurityGuard™ ULTRA F5 Cartridge: AJ0-9322

**Mobile Phase:** A: 0.1% Formic Acid in Water

B: 0.1% Formic Acid in Methanol

Gradient	Time (min)	% B
	0	80
	6	80
	6.01	100
	8	100
	8.01	80
	9.5	80

**Injection Volume:** 25 µL

**Flow Rate:** 0.3 mL/min

**Temperature:** Ambient

**Detection:** MS/MS (SCIEX Triple Quad™ 4500), APCI+

**HPLC System:** Agilent® 1100 HPLC with Quaternary Pump

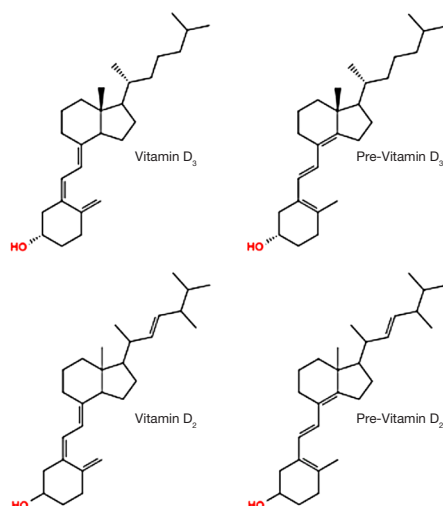
**Sample:** 1. Vitamin D<sub>2</sub>

2. Pre-Vitamin D<sub>3</sub>

3. Vitamin D<sub>3</sub>

4. Pre-Vitamin D<sub>3</sub>

### Vitamin D and Pre-Vitamin D Structures



### Total Assay Recovery

Sample ID	Recovery (%)
Vitamin D <sub>2</sub>	38.0
Vitamin D <sub>3</sub>	33.7
Pre-Vitamin D <sub>2</sub>	30.0
Pre-Vitamin D <sub>3</sub>	31.4

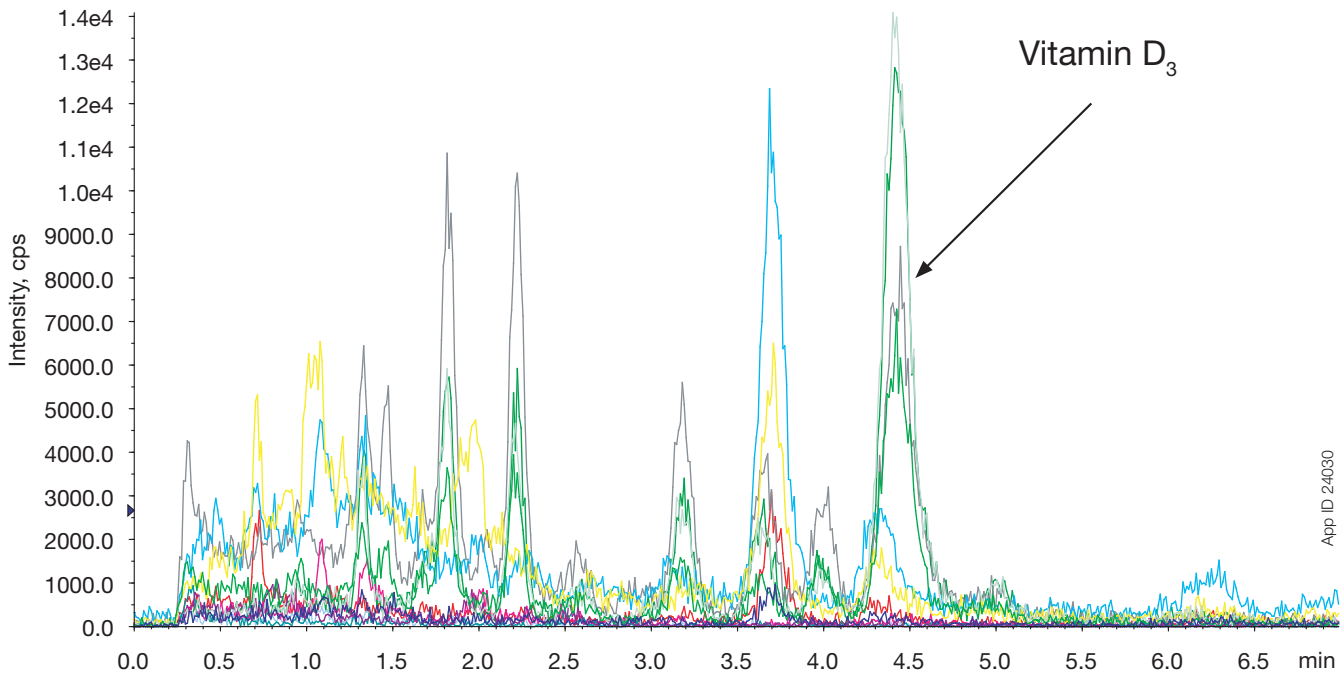
Recovery calculation includes pre-treatment, LLE and SPE extractions, resulting in average recovery for each step of near 70% for all analytes. (70% Pretreatment→70% LLE→70% SPE)

View full technical note at  
[www.phenomenex.com/foodresources](http://www.phenomenex.com/foodresources)

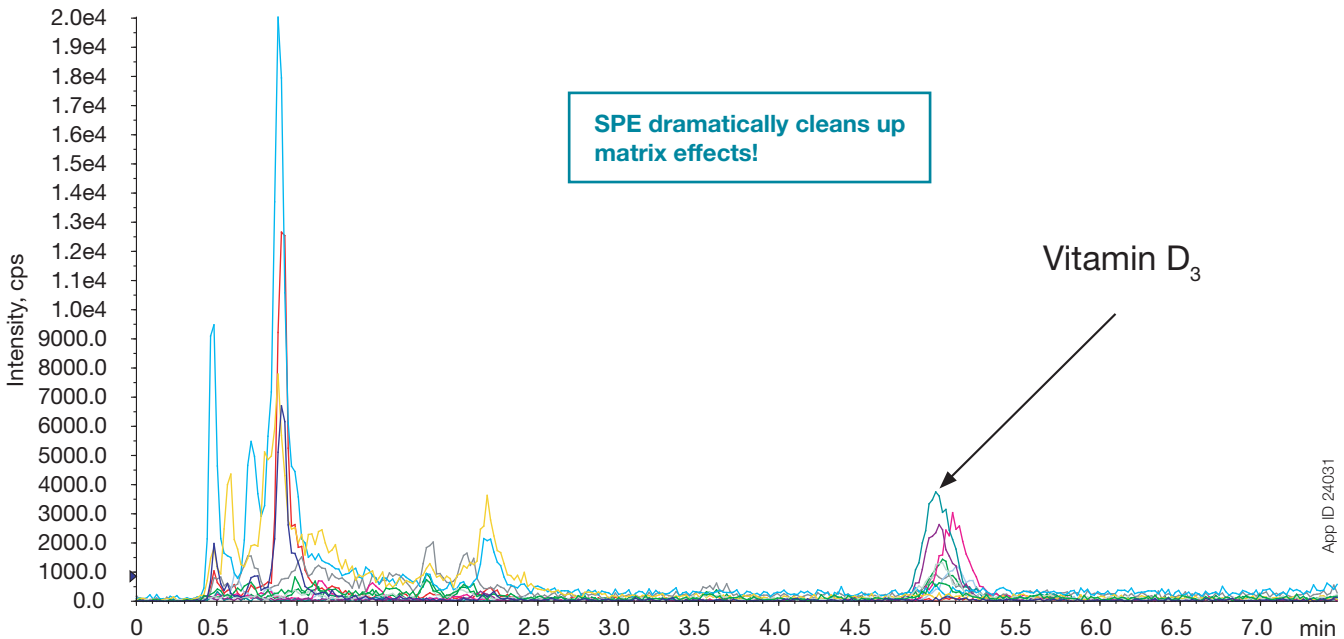
# Vitamin D2/D3 and Pre-D2/D3 in Pet Food (cont'd)

## Matrix Effects: LLE vs. LLE and SPE

### LLE Only



### LLE and SPE



# FAME Analysis in Olive Oil

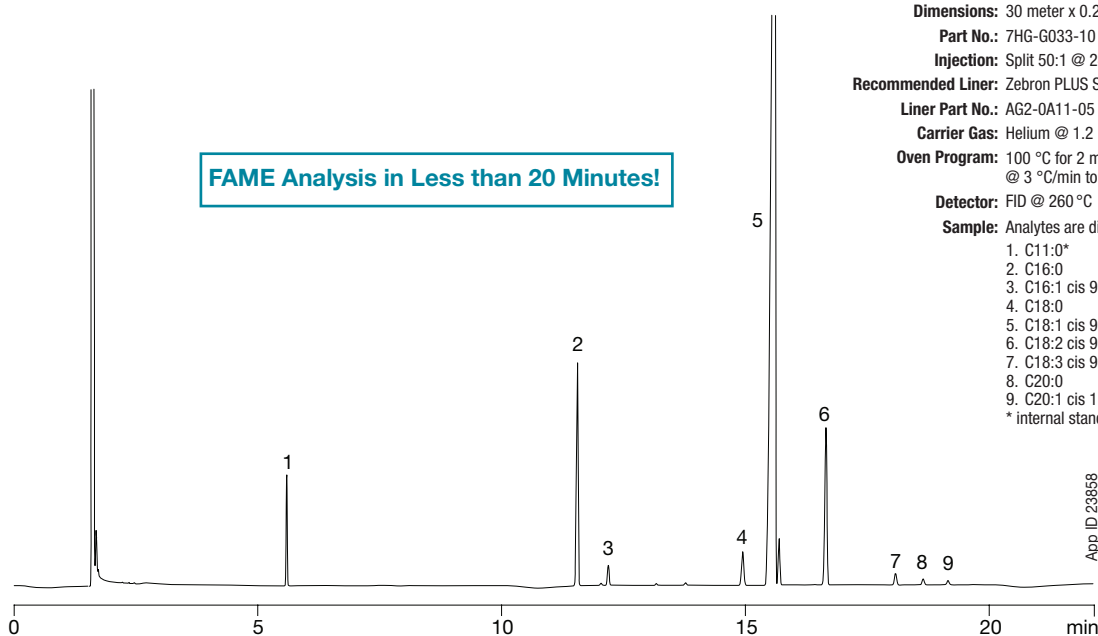
## Using Strata SPE and GC-FID

Olive oil is one of the most adulterated food products worldwide. Understanding the profile of fatty acids present in oil helps the marketplace provide an authentic and reliable product to the tables of consumers across the globe. This is why the International Olive Council created method COI/T.20/Doc. No 25, which is the global method for the detection of extraneous oils in olive oils. In this method, oil extraction is performed by Strata Si SPE and a Zebron™ ZB-FAME GC column is used to achieve good accuracy and fast run times.

### SPE Protocol

<b>Cartridge:</b>	Strata Si-1, 1g/6 mL (on vacuum or positive pressure manifold)
<b>Part No.:</b>	8B-S012-JCH
<b>Condition:</b>	6 mL Hexane
<b>Load:</b>	Oil solution (0.12 g of oil in 0.5 mL of Hexane)
<b>Elute:</b>	10 mL of Hexane/Diethyl ether (87:13)
<b>Dry Down:</b>	Evaporate eluate under a steady stream of Nitrogen
<b>Dissolve:</b>	Purified oil residue in 1 mL Hexane
<b>Add:</b>	0.1 mL 2 N Potassium hydroxide in Methanol
<b>Shake:</b>	Cap tube and shake vigorously for 15 seconds; leave to separate until upper layer becomes clear
<b>Extract:</b>	Upper layer for analysis (the heptane solution is suitable for injection into the GC)

### Extra Virgin Olive Oil FAMES



### GC-FID Conditions

<b>Column:</b>	Zebron ZB-FAME
<b>Dimensions:</b>	30 meter x 0.25 mm x 0.20 μm
<b>Part No.:</b>	7HG-G033-10
<b>Injection:</b>	Split 50:1 @ 240 °C, 1 μL
<b>Recommended Liner:</b>	Zebron PLUS Single Taper with Wool
<b>Liner Part No.:</b>	AG2-0A11-05 (for Agilent® system)
<b>Carrier Gas:</b>	Helium @ 1.2 mL/min (constant flow)
<b>Oven Program:</b>	100 °C for 2 min to 140 °C @ 10 °C/min to 190 °C @ 3 °C/min to 260 °C @ 30 °C/min for 2 min
<b>Detector:</b>	FID @ 260 °C
<b>Sample:</b>	Analytes are diluted 5:1 in heptane
	1. C11:0*
	2. C16:0
	3. C16:1 cis 9
	4. C18:0
	5. C18:1 cis 9
	6. C18:2 cis 9,12
	7. C18:3 cis 9,12,15
	8. C20:0
	9. C20:1 cis 11
	* internal standard

Find more SPE applications  
Visit [www.phenomenex.com/SPE](http://www.phenomenex.com/SPE)



# Sample Preparation Ordering Information

## roQ™ Extraction Kits

Extraction kits contain fifty easy-pour salt packets and fifty 50 mL stand-alone centrifuge tubes.

Description	Unit	Part No.
<b>AOAC 2007.01 Method Extraction Kits</b>		
6.0 g MgSO <sub>4</sub> , 1.5 g NaOAc	50/pk	KSO-8911*
<b>EN 15662 Method Extraction Kits</b>		
4.0 g MgSO <sub>4</sub> , 1.0 g NaCl, 1.0 g SCTD, 0.5 g SCDS	50/pk	KSO-8909*
<b>Original Non-buffered Method Extraction Kits</b>		
4.0 g MgSO <sub>4</sub> , 1.0 g NaCl	50/pk	KSO-8910
6.0 g MgSO <sub>4</sub> , 1.5 g NaCl	50/pk	KSO-8912

\*AOAC and EN Extraction Kits also available in traditional non-collared 50 mL centrifuge tubes, Part No.: KSO-8911-NC and KSO-8909-NC

## roQ dSPE Kits

dSPE kits contain pre-weighed sorbents/salts inside 2 mL or 15 mL centrifuge tubes.

Description	Unit	Part No.
<b>2 mL dSPE Kits</b>		
150 mg MgSO <sub>4</sub> , 25 mg PSA, 25 mg C18E	100/pk	KSO-8913
150 mg MgSO <sub>4</sub> , 25 mg PSA, 2.5 mg GCB	100/pk	KSO-8914
150 mg, MgSO <sub>4</sub> , 25 mg PSA, 7.5 mg GCB	100/pk	KSO-8915
150 mg MgSO <sub>4</sub> , 25 mg PSA	100/pk	KSO-8916
150 mg MgSO <sub>4</sub> , 50 mg PSA, 50 mg C18E, 50 mg GCB	100/pk	KSO-8917
150 mg MgSO <sub>4</sub> , 50 mg PSA, 50 mg C18E	100/pk	KSO-8918
150 mg MgSO <sub>4</sub> , 50 mg PSA, 50 mg GCB	100/pk	KSO-8919
150 mg MgSO <sub>4</sub> , 50 mg PSA	100/pk	KSO-8920
<b>15 mL dSPE Kits</b>		
900 mg MgSO <sub>4</sub> , 150 mg PSA, 150 mg C18E	50/pk	KSO-8921
900 mg MgSO <sub>4</sub> , 150 mg PSA, 15 mg GCB	50/pk	KSO-8922
900 mg MgSO <sub>4</sub> , 150 mg PSA, 45 mg GCB	50/pk	KSO-8923
900 mg MgSO <sub>4</sub> , 150 mg PSA	50/pk	KSO-8924
1200 mg MgSO <sub>4</sub> , 400 mg PSA, 400 mg C18E, 400 mg GCB	50/pk	KSO-8925
1200 mg MgSO <sub>4</sub> , 400 mg PSA, 400 mg C18E	50/pk	KSO-8926
1200 mg MgSO <sub>4</sub> , 400 mg PSA, 400 mg GCB	50/pk	KSO-8927
1200 mg MgSO <sub>4</sub> , 400 mg PSA	50/pk	KSO-8928

## Novum™ SLE

Description	Unit	Part No.
Novum SLE MINI 96-Well Plate	1/pk	8E-S138-FGA
Novum SLE MAX 96-Well Plate	1/pk	8E-S138-5GA
Novum SLE 1 cc tubes	100/pk	8B-S138-FAK
Novum SLE 3 cc tubes	50/pk	8B-S138-5BJ
Novum SLE 6 cc tubes	30/pk	8B-S138-JCH
Novum SLE 12 cc tubes	20/pk	8B-S138-KDG

## Strata® DE SLE

Description	Unit	Part No.
Strata DE SLE 200 µL 96-Well Plate	2/pk	8E-S325-FGB
Strata DE SLE 400 µL 96-Well Plate	2/pk	8E-S325-5GB
Strata DE SLE 12 cc Tube	20/pk	8B-S325-KDG
Strata DE SLE 60 cc Tube	16/pk	8B-S325-VFF

## roQ Extraction Salt Packets

Salt packets only. Centrifuge tubes not included.

Description	Unit	Part No.
<b>AOAC 2007.01 Method Extraction Packets</b>		
6.0 g MgSO <sub>4</sub> , 1.5 g NaOAc	50/pk	AHO-9043
<b>EN 15662 Method Extraction Packets</b>		
4.0 g MgSO <sub>4</sub> , 1.0 g NaCl, 1.0 g SCTD, 0.5 g SCDS	50/pk	AHO-9041
<b>Original Non-Buffered Method Extraction Packets</b>		
4.0 g MgSO <sub>4</sub> , 1.0 g NaCl	50/pk	AHO-9042
6.0 g MgSO <sub>4</sub> , 1.5 g NaCl	50/pk	AHO-9044

## Bulk roQ QuEChERS Sorbents

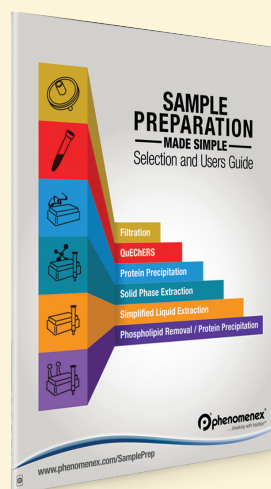
Phase	10 g	100 g
C18-E	—	04G-4348
GCB (Graphitized Carbon Black)	04D-4615	04G-4615
PSA	—	04G-4610

## Sample Preparation Selection and Users Guide

Over 50 pages to assist you in selecting and using the appropriate technique.

Request Your Copy:

[www.phenomenex.com/SPguide](http://www.phenomenex.com/SPguide)



# Sample Preparation Ordering Information



## Strata-X Polymeric SPE Sorbents

Tubes	1 mL (100/box)		3 mL (50/box)			6 mL (30/box)		
	30 mg	60 mg	60 mg	200 mg	500 mg	100 mg	200 mg	500 mg
Strata-X	8B-S100-TAK	8B-S100-UAK	8B-S100-UBJ	8B-S100-FBJ	8B-S100-HBJ	8B-S100-ECH	8B-S100-FCH	8B-S100-HCH
Strata-X-C	8B-S029-TAK	—	8B-S029-UBJ	8B-S029-FBJ	8B-S029-HBJ	8B-S029-ECH	8B-S029-FCH	8B-S029-HCH
Strata-X-CW	8B-S035-TAK	—	8B-S035-UBJ	8B-S035-FBJ	8B-S035-HBJ	8B-S035-ECH	8B-S035-FCH	8B-S035-HCH
Strata-X-A	8B-S123-TAK	—	8B-S123-UBJ	8B-S123-FBJ	8B-S123-HBJ	8B-S123-ECH	8B-S123-FCH	8B-S123-HCH
Strata-X-AW	8B-S038-TAK	—	8B-S038-UBJ	8B-S038-FBJ	8B-S038-HBJ	8B-S038-ECH	8B-S038-FCH	8B-S038-HCH
Strata-XL	8B-S043-TAK	—	8B-S043-UBJ	8B-S043-FBJ	8B-S043-HBJ	8B-S043-ECH	8B-S043-FCH	8B-S043-HCH
Strata-XL-C	8B-S044-TAK	—	8B-S044-UBJ	8B-S044-FBJ	8B-S044-HBJ	8B-S044-ECH	8B-S044-FCH	8B-S044-HCH
Strata-XL-CW	8B-S052-TAK	—	8B-S052-UBJ	8B-S052-FBJ	8B-S052-HBJ	8B-S052-ECH	8B-S052-FCH	8B-S052-HCH
Strata-XL-A	8B-S053-TAK	—	8B-S053-UBJ	8B-S053-FBJ	8B-S053-HBJ	8B-S053-ECH	8B-S053-FCH	8B-S053-HCH
Strata-XL-AW	8B-S051-TAK	—	8B-S051-UBJ	8B-S051-FBJ	8B-S051-HBJ	8B-S051-ECH	8B-S051-FCH	8B-S051-HCH

Giga™ Tubes	12mL		20 mL		60 mL
	500 mg	1g	1mg	2 mg	5 mg
Strata-X	8B-S100-HDG	8B-S100-JDG	8B-S100-JEG	8B-S100-KEG	8B-S100-LFF
Strata-X-C	8B-S029-HDG	8B-S029-JDG	8B-S029-JEG	8B-S029-KEG	8B-S029-LFF
Strata-X-CW	8B-S035-HDG	8B-S035-JDG	8B-S035-JEG	8B-S035-KEG	8B-S035-LFF
Strata-X-A	8B-S123-HDG	8B-S123-JDG	8B-S123-JEG	8B-S123-KEG	8B-S123-LFF
Strata-X-AW	8B-S028-HDG	8B-S038-JDG	8B-S038-JEG	8B-S038-KEG	8B-S038-LFF



## Strata Silica-Based SPE Sorbents

Tubes	1 mL (100/box)		3 mL (50/box)			6 mL (30/box)		
	50 mg	100 mg	100 mg	200 mg	500 mg	200 mg	500 mg	1 g
C18-E	8B-S001-DAK	8B-S001-EAK	8B-S001-EBJ	8B-S001-FBJ	8B-S001-HBJ	8B-S001-FCH	8B-S001-HCH	8B-S001-JCH
C18-U	—	8B-S002-EAK	—	8B-S002-FBJ	8B-S002-HBJ	—	8B-S002-HCH	8B-S002-JCH
C18-T	—	8B-S004-EAK	—	8B-S004-FBJ	8B-S004-HBJ	—	8B-S004-HCH	8B-S004-JCH
C8	—	8B-S005-EAK	—	8B-S005-FBJ	8B-S005-HBJ	—	8B-S005-HCH	8B-S005-JCH
Phenyl	—	8B-S006-EAK	—	8B-S006-FBJ	8B-S006-HBJ	—	8B-S006-HCH	8B-S006-JCH
SCX	—	8B-S010-EAK	8B-S010-EBJ	8B-S010-FBJ	8B-S010-HBJ	—	8B-S010-HCH	8B-S010-JCH
WCX	—	8B-S027-EAK	—	8B-S027-FBJ	8B-S027-HBJ	—	8B-S027-HCH	8B-S027-JCH
SAX	—	8B-S008-EAK	8B-S008-EBJ	8B-S008-FBJ	8B-S008-HBJ	—	8B-S008-HCH	8B-S008-JCH
NH <sub>2</sub>	—	8B-S009-EAK	—	8B-S009-FBJ	8B-S009-HBJ	—	8B-S009-HCH	8B-S009-JCH
CN	—	8B-S007-EAK	—	8B-S007-FBJ	8B-S007-HBJ	—	8B-S007-HCH	8B-S007-JCH
Si-1	—	8B-S012-EAK	—	8B-S012-FBJ	8B-S012-HBJ	—	8B-S012-HCH	8B-S012-JCH
Florisil®	—	—	—	—	8B-S013-HBJ	—	8B-S013-HCH	8B-S013-JCH
EPH	—	—	—	—	8B-S031-HBJ	—	—	—
AL-N	—	—	—	—	8B-S313-HBJ	—	—	8B-S313-JCH

Giga Tubes	12mL		20 mL	60 mL	
	500 mg	1g	5mg	5 mg	20 mg
Strata NH <sub>2</sub>	8B-S009-HDG	8B-S009-KDG	8B-S009-LEG	8B-S009-MFF	8B-S009-VFF
Strata Alumina	8B-S313-HDG	8B-S313-JDG	8B-S313-LEG	8B-S313-MFF	—

## Presston 100 Positive Pressure Manifold

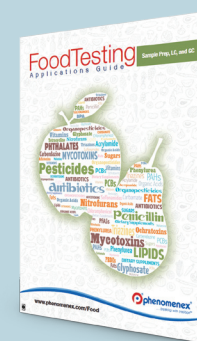
Part No.	Description	Unit
AHO-9342	Presston 100 Positive Pressure Manifold, 1 mL Tube Complete Assembly	1/Box
AHO-9347	Presston 100 Positive Pressure Manifold, 3 mL Tube Complete Assembly	1/Box
AHO-9343	Presston 100 Positive Pressure Manifold, 6 mL Tube Complete Assembly	1/Box



Phenomenex warrants that for a period of 12 months following delivery, the Presston 100 Positive Pressure Manifold you have purchased will perform in accordance with the published specifications and will be free from defects in materials or workmanship. In the event that the Presston 100 Positive Pressure Manifold does not meet this warranty, Phenomenex will repair or replace defective parts. Please visit [www.phenomenex.com/Presston](http://www.phenomenex.com/Presston) for complete warranty information.



## Comprehensive Food Testing Guide



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[www.phenomenex.com/FTGuide](http://www.phenomenex.com/FTGuide)

# LC Ordering Information

## Kinetex Core-Shell HPLC Columns

Kinetex 5 µm Columns (mm)	SecurityGuard™ ULTRA Cartridges†		SecurityGuard™ ULTRA Cartridges†				SecurityGuard™ ULTRA Cartridges†		
	50 x 2.1	3/pk	50 x 3.0	3/pk	50 x 4.6	100 x 4.6	150 x 4.6	250 x 4.6	3/pk
Biphenyl	00B-4627-AN	AJ0-9209	00B-4627-Y0	AJ0-9208	00B-4627-E0	00D-4627-E0	00F-4627-E0	00G-4627-E0	AJ0-9207
		for 2.1 mm ID		for 3.0 mm ID					for 4.6 mm ID

2.6 µm Minibore Columns (mm)						SecurityGuard ULTRA Cartridges†
Phases	30 x 2.1	50 x 2.1	75 x 2.1	100 x 2.1	150 x 2.1	3/pk
Biphenyl	00A-4622-AN	00B-4622-AN	---	00D-4622-AN	00F-4622-AN	AJ0-9209
XB-C18	00A-4496-AN	00B-4496-AN	00C-4496-AN	00D-4496-AN	00F-4496-AN	AJ0-8782
F5	00A-4723-AN	00B-4723-AN	---	00D-4723-AN	00F-4723-AN	AJ0-9322

for 2.1 mm ID



†SecurityGuard ULTRA Cartridges required holder, Part No.: AJ0-9000.

More Kinetex Phases dimensions available at [www.phenomenex.com/Kinetex](http://www.phenomenex.com/Kinetex)

## Synergi™ Hydro-RP HPLC Columns

4 µm Microbore and Minibore Columns (mm)									SecurityGuard Cartridges*
Phases	50 x 1.0	150 x 1.0	250 x 1.0	30 x 2.0	50 x 2.0	75 x 2.0	150 x 2.0	250 x 2.0	4 x 2.0*
Hydro-RP	00B-4375-A0	00F-4375-A0	00G-4375-A0	00A-4375-B0	00B-4375-B0	00C-4375-B0	00F-4375-B0	00G-4375-B0	AJ0-7510
									for ID: 2.0-3.0 mm

\*SecurityGuard Analytical Cartridges require holder, Part No.: KJ0-4282

# GC Ordering Information

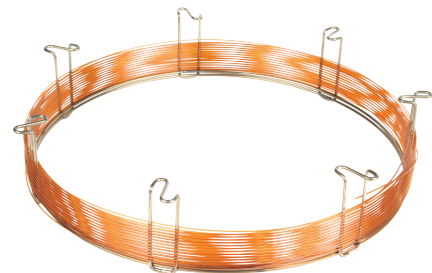
## Zebbron ZB-FAME GC Columns

ID (mm)	df (µm)	Temp. Limits °C	Part No.	5 m Guardian
<b>20-Meter</b>				
0.18	0.15	-20 to 280	7FD-G033-05	---
<b>30-Meter</b>				
0.25	0.20	-20 to 280	7HG-G033-10	7HG-G033-10-GGA
<b>60-Meter</b>				
0.25	0.20	-20 to 280	7KG-G033-10	---



## Zebbron ZB-5<sup>PLUS</sup>™ GC Columns

ID (mm)	df (µm)	Temp. Limits (°C)	Part No.	5 m Guardian
<b>30 Meter</b>				
0.25	0.25	-60 to 360/370	7HG-G032-11	7HG-G032-11-GGA
0.25	0.50	-60 to 360/370	7HG-G032-17	---
0.25	1.00	-60 to 360/370	7HG-G032-22	---
0.32	0.25	-60 to 360/370	7HM-G032-11	---
0.32	1.00	-60 to 360/370	7HM-G032-22	---



## Zebbron ZB-MultiResidue™ GC columns (MR)-1

ID (mm)	df (µm)	Temp. Limits °C	Part No.
<b>20-Meter</b>			
0.18	0.18	-60 to 320/340	7FD-G016-08
<b>30-Meter</b>			
0.25	0.25	-60 to 320/340	7HG-G016-11
0.32	0.25	-60 to 320/340	7HM-G016-11
0.32	0.50	-60 to 320/340	7HM-G016-17
0.53	0.50	-60 to 320/340	7HK-G016-17

## Zebbron ZB-MultiResidue GC columns (MR)-2

ID (mm)	df (µm)	Temp. Limits °C	Part No.
<b>30-Meter</b>			
0.25	0.20	-60 to 320/340	7HG-G017-10
0.32	0.25	-60 to 320/340	7HM-G017-11
0.53	0.50	-60 to 320/340	7HK-G017-17

Note: If you need a 5 in. cage, simply add a (-B) after the part number, e.g., 7HG-G016-11-B or 7HG-G017-10-B. Some exceptions may apply. Agilent 6850 and some SRI and process GC systems use only 5 in. cages.



If Phenomenex products in this brochure do not provide at least an equivalent separation as compared to other products of the same phase and dimensions, return the product with comparative data within 45 days for a FULL REFUND.

# FOOD TESTING SAMPLE PREPARATION

IMPROVE | CLEAN | SAVE

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