Supel™ QuE Sample Preparation

Supelco®
Analytical Products

Supel™ QuE Sample Preparation

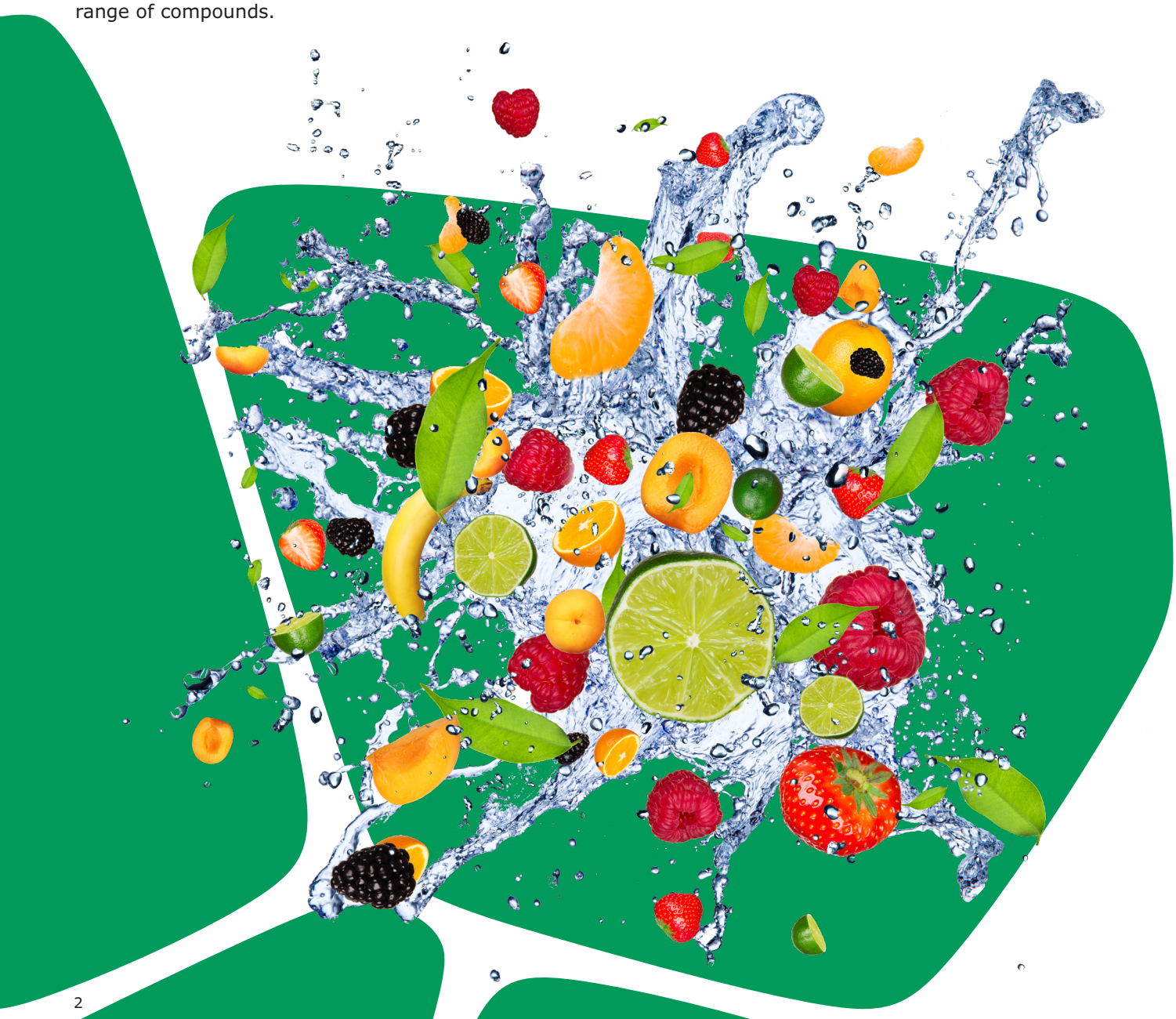
Multi-residue pesticide analysis of food and agricultural samples using QuEChERS

In “QuEChERS” methodology, the use of loose extraction salts and cleanup sorbents in combination with shaking and centrifugation results in a Quick, Easy, Cheap, Effective, Rugged and Safe sample cleanup technique. The “QuEChERS” method has emerged as a sample prep technique popular for multi-residue pesticide analysis in food and agricultural products, and is formalized in method EN15662:2008 and AOAC 2007.01.[1-2] Recently, QuEChERS has been expanded to include the analysis of PAH, PCB, PBDE and flame retardants analysis.[3]

The Supel™ QuE line of centrifuge tubes contains predetermined amounts of salts and SPE sorbents to support the most common method configurations used today for QuEChERS.

Features and Impact

QuEChERS significantly improves laboratory efficiency and throughput. This procedure requires only small quantities of solvent and is capable of generating recoveries of 70-120% with RSDs <5% for a wide range of compounds.



A PROCESS IN TWO STEPS

1-Extraction

- Solvent extraction techniques are designed to achieve maximum yield of analytes from the base matrix
- Solvent selection is important to minimize co-extracting compounds
- Analytes are extracted from the matrix with acetonitrile and salts/buffers

2-Cleanup

- Sample cleanup is necessary to reduce interferences
- Interferences can damage analytical instrumentation and complicate analyte identification and quantification
- The use of PSA, C18, GCB (Graphitized Carbon Black) or Z-Sep sorbents allow removal of sugars, lipids, sterols, organic acids, proteins, carotenoids, chlorophyll and other pigments prior to a GC-MS/MS or LC-MS/MS analysis

OFFICIAL QUECHERS METHODS

EN 15662:2008

- The European method includes sodium chloride to limit polar interferences and several buffering citrate based reagents to preserve base-sensitive analytes
- The use of sodium hydroxide in the citrate step should be avoided as it damages the sorbent used in the cleanup step

AOAC 2007.01

- Employs 1% acetic acid in acetonitrile and sodium acetate buffer to protect base sensitive analytes from degradation
- A USDA study has demonstrated that this method provides superior recovery for pH sensitive compounds when compared to the other QuEChERS methods. Since the approach uses acetic acid in the extraction step, the PSA sorbent used in the cleanup step might get overloaded making it less effective in cleanup and possibly causing GC resolution/ backgound issues.

EN 15662:2008	AOAC 2007.01
Freeze samples to -20 °C. Homogenize with dry ice until a free flowing powder is formed.	Freeze samples to -20 °C. Homogenize with dry ice until a free flowing powder is formed.
Step 1 - Sample Extraction	
Weigh 10 g sample into a 50 mL centrifuge tube.	Transfer 10-15 g homogenized food sample to 50 mL PTFE centrifuge tube.
Add 10 mL acetonitrile (ACN) + 100 µL internal standard (IS) solution. Shake vigorously for 1 minute.	For each 15 g sample, add 15 mL 1% acetic acid in ACN + contents of Supel™ QuE Acetate (AC) Tube (55234-U) + 75 µL I.S. solution.
Add contents of Supel™ QuE Citrate Extraction Tube (55227-U). Shake for 1 minute and centrifuge for 5 minutes at 3000 U/min.	Shake for 1 minute and centrifuge for 1 minute at 1500 U/min.
Step 2 - Cleanup	
Transfer an aliquot of the ACN layer (supernatant) to a dispersive cleanup tube. Shake for 30 seconds (2 minutes if ENVI-Carb™) Centrifuge 5 minute at 3000 U/min.	Transfer an aliquot of the ACN layer (supernatant) to a dispersive cleanup tube. Shake for 30 seconds (2 minutes if ENVI-Carb™) Centrifuge 1 minute at 1500 U/min
Acidify an aliquot of supernatant with formic acid 5% in ACN (10 µL to every mL of supernatant isolated).	Further processing may be necessary prior to chromatographic analysis (addition of formic acid, evaporation and reconstitution with toluene).

QUECHERS: BEYOND THE BASICS

QuEChERS was originally designed for fruits and vegetables and proven to work well especially for high water content fruits and vegetables (> 80%) under slightly acidic conditions (pH 5-6).[1–4] However, there are commodities that do not fit into this food type. Adjustments to the typical QuEChERS procedures extend its usage outside of these typical or easy commodities.

To improve the extraction efficiency of low moisture containing commodities, addition of water to the samples prior to extraction is recommended (see table to the right).

To reduce co-extracted materials from acidic foods,[5] buffering is needed. The buffering of the EN method is intended to produce this pH 5-6 condition during extraction.[3] The buffering capacity of existing methods cannot adequately correct for the low pH of citrus fruits (with pH < 3). In this case the EN method calls for addition of *5 N (or M, mol/L)* sodium hydroxide solution to the salt mixes. In case of lemons and limes, 600 µL are recommended.[3] For commodities like raspberry, addition of 200 µL to the extraction tube is recommended. Acidic foods outside the citrus family can be pH adjusted and generally produce comparable high-quality data as expected with non acidic foods.[1–4]

Sample Types	Sample Weight	Water Added
Fruits and vegetables > 80% water content	10 g	—
Fruits and vegetables 25-80% water content	10 g	x g
Cereals	5 g	10 g
Dried fruits	5 g	7.5 g
Honey	5 g	10 g
Spices	2 g	10 g

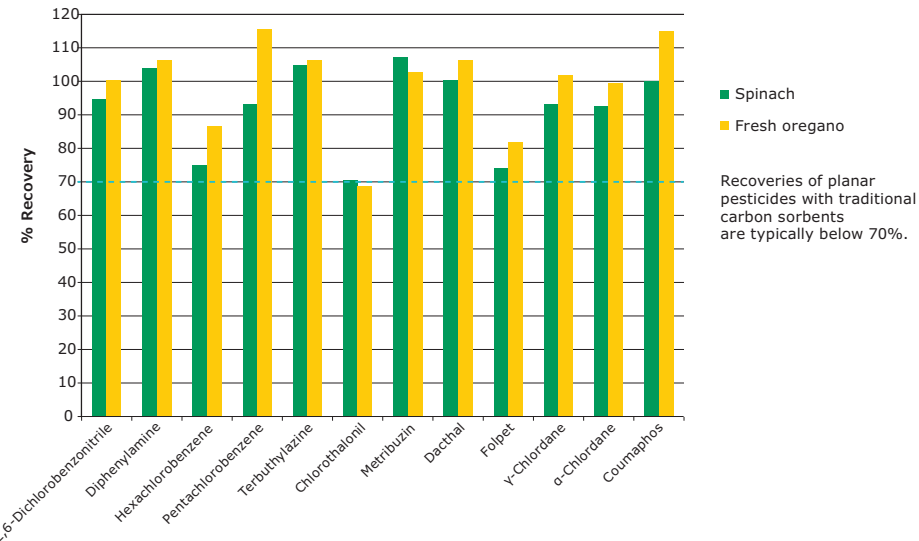
References

1. Fast and Easy Multiresidue Method Employing Acetonitrile Extraction/ Partitioning and "Dispersive Solid-Phase Extraction" for the Determination of Pesticide Residues in Produce. (M. Anastassiades, S.J. Lehotay, D. Stajnbaher, F.J. Schenck, J. AOAC International 86 (2003) 412).
2. Pesticide Residues in Foods by Acetonitrile Extraction and Partitioning with Magnesium Sulfate. (AOAC Official Method 2007.01).
3. Foods of Plant Origin—Determination of Pesticide Residues Using GC-MS and/or LC-MS/MS Following Acetonitrile Extraction/ Partitioning and Clean-up by Dispersive SPE (QuEChERS-method). (EN 15662 Version 2008).
4. Comparison of QuEChERS sample preparation methods for the analysis of pesticide residues in fruits and vegetables. (S. Lehotay, K. Son, H. Kwon, U. Koesukwiwat, W. Fu, K. Mastovska, E. Hoh, N. Leepipatpiboon, J. Chromatogr. A, 1217 (2010) 2548).
5. The QuEChERS Method –Background Information and Recent Developments, Community Reference Laboratory Pesticide Residues using Single Residue Methods. (M. Anastassiades, 1st Joint CRL-Workshop –Stuttgart, presentation (2006)).

Supel™ QuE Verde for Challenging Compounds in Green Matrices

Supel™ QuE Verde for QuEChERS combines a novel carbon with zirconia coated silica (Z-Sep+) to provide an optimum balance between analyte recovery and color removal. This sorbent combination has been shown to provide recoveries in the range of 70% to 120% of even the most challenging planar pesticides while maintaining >95% pigment removal in high chlorophyll matrices.

Supel™ QuE Verde is a mixture of an improved graphitized carbon black (GCB), Z-Sep+, and primary-secondary amine (PSA). The improved GCB has been optimized to balance chlorophyll removal and improve recoveries of planar pesticides. Z-Sep+ is a silica that is functionalized with both zirconia and C18. Zirconia will retain some fats and carotenoids, while C18 retains hydrophobic interferences. The PSA in the mix functions to remove acidic interferences. When used to clean samples containing chlorophyll, this sorbent blend will provide better recovery of planar pesticides than sorbents containing traditional GCB.



Recoveries of planar pesticides with traditional carbon sorbents are typically below 70%.

Average Percent Recoveries From Spinach and Oregano Extracts Spiked at 50 ng/mL After Cleanup With the 2 mL Supel™ QuE Verde Tube (n=3).

Discover Supel™ QuE Verde at
SigmaAldrich.com/verde

Classification of Fruits and Vegetables (as defined in the CEN Guidelines)

Commodity Groups	Typical Commodity Categories	Typical Representative Commodities
1. High water content 	Pome fruit	Apples, pears
	Stone fruit	Apricots, cherries, peaches,
	Other fruit	Bananas
	Alliums	Onions, leeks
	Fruiting vegetables/cucurbits	Tomatoes, peppers, cucumber, melon
	Brassica vegetables	Cauliflower, Brussels-sprouts, cabbage, broccoli
	Leafy vegetables and fresh herbs	Lettuce, spinach, basil
	Stem and stalk vegetables	Celery, asparagus
	Forage/fodder crops	Fresh alfalfa, fodder vetch, fresh sugar beets
	Fresh legume vegetables	Fresh peas with pods, peas, mange tout, broad beans, runner beans, French beans
	Leaves of root and tuber vegetables	Sugar beet and fodder beet tops
	Fresh fungi	Champignons, canterelles
	Root and tuber vegetables or feed	Sugar beet and fodder beet roots, carrots, potatoes, sweet potatoes
2. High acid content and high water content[1] 	Citrus fruit	Lemons, mandarins, tangerines, oranges
	Small fruit and berries	Strawberry, blueberry, raspberry, black currant, red currant, white currant, grapes
	Other	Kiwi fruit, pineapple, rhubarb
3. High sugar and low water content[2] 	Honey, dried fruit	Honey, raisins, dried apricots, dried plums, fruit jams
4a. High oil content and very low water content 	Tree nuts	Walnuts, hazelnuts
	Oil seeds	Oilseed rape, sunflower, cotton-seed, soybeans, peanuts, sesame, etc.
	Pastes of tree nuts and oil seeds	Peanut butter, tahina, hazelnut paste
	Oils from tree nuts, oil seeds and oily fruits	Olive oil, rapeseed oil, sunflower oil, pumpkin seed oil
4b. High oil content and intermediate water content 	Oily fruits and products	Olives, avocados and pastes thereof
5. High starch and/or protein content and low water and fat content 	Dry legume vegetables/pulses	Field bean, dried broad bean, dried haricot bean (yellow, white/navy, brown, speckled), lentils
	Cereal grain and products thereof	Wheat, rye, barley and oat grain; maize, rice, wholemeal bread, white bread, crackers, breakfast cereals, pasta
6. Difficult or unique commodities 	—	Hops, cocoa beans and products thereof, coffee, tea spices

[1] If a buffer is used to stabilize the pH changes in the extraction step, then commodity Group 2 can be merged with commodity Group 1.

[2] Where commodities of Group 3 are mixed with water prior to extraction to achieve a water content of >70%, this commodity group may be merged with Group 1. The RLs should be adjusted to account for smaller sample portions (e.g. if 10 g portions are used for commodities of Group 1 and 5 g for Group 3, the RL of Group 3 should be twice the RL of Group 1 unless a commodity belonging to Group 3 is successfully validated at a lower level).

HOW TO CHOOSE AN SPE SORBENT?

Supelco® centrifuge tubes are available in bulk quantities, each packed with predetermined amounts of salts and SPE sorbents to support the most common method configurations used today. This saves the analyst the time taken to weigh out the individual materials multiple times.

Cleanup Products and Their Characteristics

Zirconia on Silica (Z-Sep, Z-Sep/C18, Z-Sep+)

- Lewis Acid-Lewis base interactions between hydroxyl groups and Zirconium
- Hydrophobic interactions between triglycerides and C18 (Z-Sep/C18, Z-Sep+)
- Significantly diminishes fatty matrix interferences and various pigments
- Provides more robust LC-MS and GC-MS methods by eliminating problematic matrix interferences
- Can replace C18 and PSA phases in current methods without additional method development

Primary Secondary Amine (PSA)

- Polymerically bonded, ethylenediamine-N-propyl phase that contains both primary and secondary amines
- A weak anion exchanger with a pKa of 10.1 and 10.9
- Similar to aminopropyl SPE phases (NH_2) in terms of selectivity, but has a much higher capacity due to presence of secondary amine (0.98-1.05 mEq/g)
- Has been shown to significantly reduce matrix-enhancement effects encountered during the GC analysis of food products
- Bidendate nature of ligands allows for chelation

ENVI-Carb™

- Extreme affinity for organic polar and nonpolar compounds with ringstructures/conjugated double bonds from both nonpolar and polar matrices when used under reversed-phase conditions
- Carbon surface comprised of hexagonal ring structures, interconnected and layered into graphitic sheets
- Non-porous; nature of the carbon phase allows for rapid processing; adsorption does not require analyte diffusion into pores

Zirconia

- Removes pigments and lipids or fats (content below 15% fats
  use Z-Sep/C18, for above 15% use Z-Sep+, for hydrophobic analytes use Z-Sep)

PSA

- Used in the removal of sugars and fatty acids, organic acids, lipids and polar pigments
- When used in combination with C18, additional lipids and sterols can be removed
- When used in combination with GCBs, higher amounts of pigments can be removed

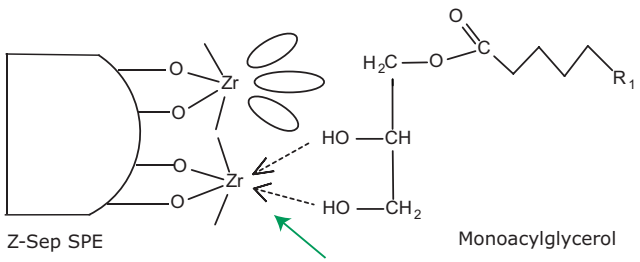
ENVI-Carb™

- Strong sorbent for removing pigments, polyphenols, and other polar compounds

C18

- Removes long carbon chain/fatty compounds, sterols and other nonpolar interferences

Retention Mechanism for Fats on Supel™ QuE Z-Sep Sorbents



Lewis acid-base interactions

Why Choose Supel™ QuE?

We were among the first to introduce QuEChERS to the market

- Excellent knowledge of the technique and manufacturing
- Extended choice of adsorbents and tube sizes (incl. 15 mL for automated shaker)

Innovative new adsorbents with unique properties

- Z-Sep sorbents for fatty and difficult food
- Supel™ QuE Verde for planar pesticides in green matrices

$MgSO_4$

- We heat the $MgSO_4$ to 550 °C prior to packing to ensure dryness

Customization Services

- Adsorbent mixes and configuration for special requirements
- Hardware options (PP containers, glass vials)
- Components to be packed (salts, adsorbents)

Did you know . . .

The Supelco® pesticide reference materials product line is the most comprehensive portfolio available in the market. Under the well-known brands PESTANAL® and *Trace*CERT®, we proudly offer more than 1300 high purity pesticide and pesticide metabolite standards and certified reference materials for food and environmental analysis including:

- TraceCERT® Certified Reference Materials (CRMs)
- CRM matrix reference materials
- Isotope labeled reference materials
- Pesticide metabolite reference materials

We add new products to the portfolio regularly, in order to keep up-to-date with new developments in pesticide analysis.

For more information, visit
SigmaAldrich.com/pesticides

SUPEL™ QUE PRODUCTS FOR QUECHERS AND RELATED PRODUCTS

Products for the Extraction Step

Supel™ QuE Tubes for the Extraction Step

Description	Qty	Cat. No.
Method EN15662 :2008 (12 mL centrifuge tubes)		
Supel™ QuE Citrate (EN) Tube – 4 g $MgSO_4$, 1 g NaCl, 0.5 g NaCitrate dibasic sesquihydrate, 1 g NaCitrate tribasic dihydrate	50	**55227-U**
Supel™ QuE Citrate/Sodium Bicarbonate (EN) Tube – 4 g $MgSO_4$, 5 g NaBicarbonate, 1 g NaCl, 0.5 g NaCitrate dibasic	50	**55237-U**
Method AOAC 2007.01 (12 mL centrifuge tubes)		
Supel™ QuE Acetate (AC) Tube – 6 g $MgSO_4$, 1.5 g NaAcetate	50	**55234-U**
Non buffered extraction tubes (12 mL centrifuge tubes)		
Supel™ QuE Non-Buffered Tube 1 – 4 g $MgSO_4$, 1 g NaCl	50	**55294-U**
Supel™ QuE Non-Buffered Tube 2 – 6 g $MgSO_4$, 1.5 g NaCl	50	**55295-U**

QuEChERS Shakers and Accessories

Description	Qty	Cat. No.
Benchmark Benchmixer™ XL Laboratory Shakers		
QuEChERS Shaker and Rack Starter Kit, USA compatible plug, AC input 115 V	—	**55278-U**
QuEChERS Shaker and Rack Starter Kit, EU compatible, schuko plug, AC input 230 V	—	**55438-U**
Multi-tube Vortexer, USA compatible plug, AC input 115 V (Sigma-Aldrich)	1	**Z765503**
Multi-tube Vortexer, EU compatible Schuko plug, AC input 230 V (Sigma-Aldrich)	—	**Z765511**
Benchmark Benchmixer XL Laboratory Shaker Racks		
50 mL QuEChERS Extraction Tube Shaker Rack	1	**55279-U**
15 mL QuEChERS Cleanup Tube Shaker Rack (Sigma-Aldrich)	1	**Z765589**
2 mL QuEChERS Cleanup Tube Shaker Rack (Sigma-Aldrich)	1	**Z765554**

Related Products

Description	Qty	Cat. No.
Ascentis® Express HPLC Columns (2.7 µm particles)		
RP-Amide, 10 cm × 2.1 mm I.D.	1	**53913-U**
Capillary GC Columns		
SLB®-5ms, 30 m × 0.25 mm I.D. × 0.25 µm	1	**28471-U**

Products for Cleanup (QuEChERS) Step

Method – EN15662:2008

Matrices*		Product Description	Cat. No. 2 mL Tubes	Cat. No. 15 mL Tubes
Groups 1, 2 and 3	General fruits and vegetables *lightly pigmented*	150 mg Supelclean™ PSA, 900 mg $MgSO_4$	—	**55437-U**
		25 mg Supelclean™ PSA, 150 mg $MgSO_4$	**55172-U**	—
	Pigmented fruits and Vegetables *Moderate levels of chlorophyll and carotenoids*	150 mg Supelclean™ PSA, 15 mg Supelclean™ ENVI-Carb™, 900 mg $MgSO_4$	—	**55446-U**
		25 mg Supelclean™ PSA, 2,5 mg Supelclean™ ENVI-Carb™, 150 mg $MgSO_4$	**55147-U**	—
	Pigmented fruits and vegetables *High levels of chlorophyll and carotenoids*	150 mg Supelclean™ PSA, 45 mg Supelclean™ ENVI-Carb™, 900 mg $MgSO_4$	—	**55464-U**
		25 mg Supelclean™ PSA, 7,5 mg Supelclean™ ENVI-Carb™, 150 mg $MgSO_4$	**55176-U**	—
Groups 4, 5 and 6	Pigmented fruits and vegetables with waxes/lipids High Lipid Content	150 mg Supelclean™ PSA, 150 mg Discovery® DSC-18, 900 mg $MgSO_4$	—	**55439-U**
		25 mg Supelclean™ PSA, 25 mg Discovery® DSC-18, 150 mg $MgSO_4$	**55173-U**	—

Method – AOAC 2007.01

Matrices*		Product Description	Cat. No. 2 mL Tubes	Cat. No. 15 mL Tubes
Groups 1, 2 and 3	General fruits and vegetables *lightly pigmented*	400 mg Supelclean™ PSA, 1200 mg $MgSO_4$	—	**55466-U**
		50 mg Supelclean™ PSA, 150 mg $MgSO_4$	**55287-U**	—
	Pigmented fruits and vegetables *Moderate levels of chlorophyll and carotenoids*	50 mg Supelclean™ PSA, 150 mg $MgSO_4$, 50 mg ENVI-Carb™	**on request**	—
	Pigmented fruits and Vegetables *High levels of chlorophyll and carotenoids*	50 mg Supelclean™ PSA, 150 mg $MgSO_4$, 50 mg Discovery® DSC-18, 50 mg ENVI-Carb™	**55289-U**	—
		400 mg Supelclean™ PSA, 1200 mg $MgSO_4$, 400 mg Discovery® DSC-18, 400 mg ENVI-Carb™		**55474-U**
Groups 4, 5 and 6	Pigmented fruits and vegetables with waxes/lipids High Lipid Content	50 mg Supelclean™ PSA, 150 mg $MgSO_4$, 50 mg Discovery® DSC-18	**55288-U**	—
		400 mg Supelclean™ PSA, 1200 mg $MgSO_4$, 400 mg Discovery® DSC-18	—	**55470-U**

Specialty Products for Method EN15662 or AOAC 2007.01 – Alternatives

Matrices*		Product Description	Cat.No. 2 mL tubes	Cat.No. 15 mL tubes
Groups 4, 5 and 6	Fatty or pigmented matrix *With less than 15% fat*	20 mg Z-Sep, 50 mg Discovery® DSC-C18 (alternative)	**55284-U**	—
		120 mg Z-Sep, 300 mg C18 (alternative)	—	**55506-U**
	Hydrophobic analytes in fatty matrices	75 mg Z-Sep (alternative)	**55411-U**	—
		50 mg Z-Sep, 150 mg $MgSO_4$ (alternative)	**55417-U**	—
		500 mg Z-Sep, (alternative)	—	**55491-U**
		300 mg Z-Sep, 900 mg $MgSO_4$ (alternative)	—	**55503-U**
	Fatty matrices *With greater than 15% fat*	75 mg Z-Sep+ (alternative)	**55408-U**	—
		50 mg Z-Sep+, 150 mg $MgSO_4$ (alternative)	**55414-U**	—
		500 mg Z-Sep+, (alternative)	—	**55486-U**
		300 mg Z-Sep+, 900 mg $MgSO_4$ (alternative)	—	**55511-U**
	Improved recovery of *planar pesticides* in green matrices	Supel™ QuE Verde, 150 mg $MgSO_4$, 10 mg ENVI™-Carb Y, 50 mg PSA, 60 mg Z-Sep+	**55447-U**	—
		Supel™ QuE Verde, 1200 mg $MgSO_4$, 80 mg ENVI™-Carb Y, 400 mg PSA, 480 mg Z-Sep+	—	**55442-U**

*See page 5 for group definitions.

APPLICATIONS

Intralab Validation of the EN 15662 Method for the Determination of 200 Pesticide Residues Using a Fused-Core® Ascentis® Express RP-Amide HPLC Column by LC-MS/MS and Cleanup by Dispersive SPE (QuEChERS)

Enio Belotti, Luca Meni, Marco Ruggeri, Water and Life Entratico (BG) Italy

Experimental

The European guideline SANCO/3131/2007 document (Method Validation and Quality Control Procedures for Pesticide Residues Analysis in Foods and Feeds) was followed for the following different representative fruit and plant origin matrices:

- Pear as sugar matrix
- Kiwi as acid matrix
- Lettuce as chlorophyll matrix
- Maize flour as cereal

The QuEChERS Multi-residue method described in EN 15662 was used to prepare the final samples to be injected into the LC/MS/MS system.

Chromatography

The HPLC method developed here utilizes an Ascentis® Express RP-Amide column as an alternative to the C18 column traditionally used in the EN 15662 method.

Combining an embedded polar group (EPG) stationary phase with the Fused-Core® particles, the Ascentis® Express RP-Amide column provides a host of useful benefits to the HPLC chromatographer.

The benefits come from both the phase technology and the particle technology, and can be summarized as:

Fused-Core® Benefits

- Twice the efficiency of traditional 3 µm HPLC columns
- Half the backpressure of sub-2 micron columns
- Capable of UHPLC performance on traditional HPLC systems

RP-Amide Benefits

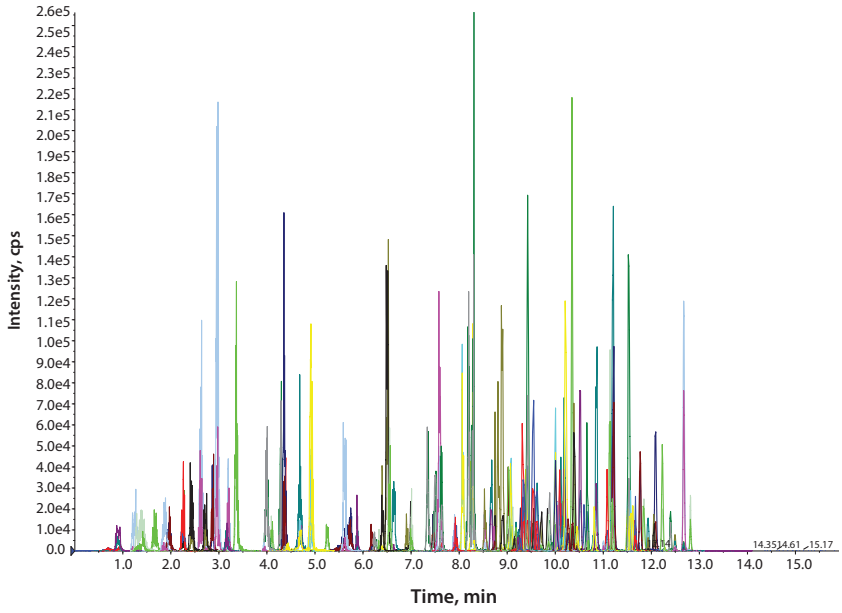
- Alternative reversed-phase selectivity to C18
- Improved peak shape for bases
- 100% aqueous compatible reversed-phase column

Alternative Selectivity

The Ascentis® Express RP-Amide column provides increased selectivity for polar compounds, especially those that can act as a hydrogen-bond donor. The selectivity differences between the RP-Amide and the C18 can be a useful tool in method development. In many cases, when peaks co-elute on a C18 phase, the RP-Amide can be substituted to achieve separation without a change in mobile phase.

column:	Ascentis® Express RP-Amide, 10 cm × 2.1 mm ID, 2.7 µm particle size
HPLC:	Shimadzu Prominence UFLC XR
MS/MS detector:	Applied Biosystems API 3200
mobile phase A:	Ammonium formate solution in water (5 mmol/l, 0.1% formic acid)
mobile phase B:	Ammonium formate solution in methanol (5 mmol/l, 0.1% formic acid)
column temp:	40 °C
injection volume:	2 µL
elution:	gradient

Time	Mobile Phase A %	Mobile Phase B %
0	95	5
0.5	90	10
12	5	95
15	85	95



For more details, download the Reporter Europe Vol. 39 /2010 at **SigmaAldrich.com/thereporter**

Determination of 113 Pesticide Residues in High Oil Vegetal Commodities

Łukasz Rajski, Ana Lozano, Ana Uclés, Carmen Ferrer, Amadeo R. Fernández-Alba - J. Chromatogr. A 1304 (2013) 109– 120

Prof. Fernandez-Alba* and his team have evaluated several extraction methods in terms of recoveries and extraction precision for 113 pesticides in avocado including QuEChERS with various dSPE cleanups, miniLuke, and ethyl acetate. This work is described in a paper published in Journal of Chromatography A.

Supel™ QuE Z-Sep and Z-Sep+, two sorbents containing ZrO_2, were used to improve fat removal from the extracts. The findings were that the QuEChERS protocol with Z-Sep cleanup showed the highest number of pesticides with recoveries in the 70–120% range, along with the lowest amount of co-extracted matrix compounds for avocado and almond matrices. As part of method validation, recoveries at two levels (10 and 50 µg/kg), limit of quantitation, linearity, matrix effects, as well as the inter- and intra-day precision were studied.

* Pesticide Residue Research Group, European Union Reference Laboratory (EURL), Department of Hydrogeology and Analytical Chemistry, University of Almería, Ctra. Sacramento S/N', La Cañada de San Urbano, 04120, Almería, Spain

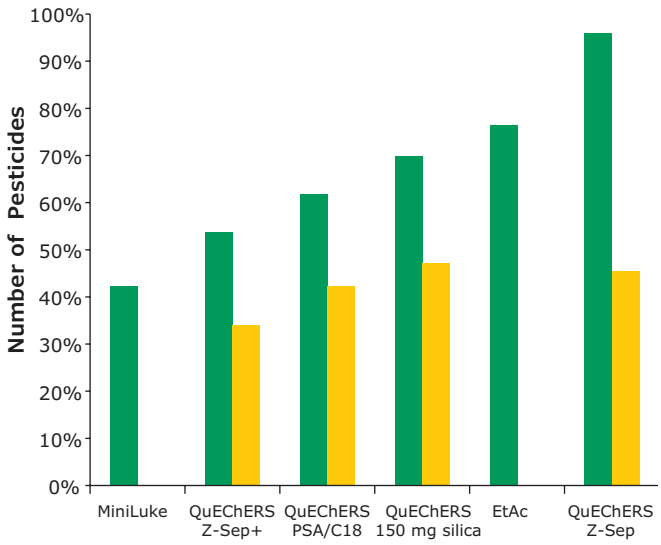
In addition to providing the best recoveries, Z-Sep cleaned extracts also exhibited the lowest average % RSD values.

The authors also showed that apart from the application of a suitable sorbent, the method for sample reconstitution after evaporation (if applied) is very important. Reconstitution in a mixture of 1:9 (v/v) acetonitrile:water was inefficient. In avocado, in order to achieve good recoveries for a large number of pesticides, it was necessary to increase the amount of acetonitrile to 30%. Almond samples, on the other hand, required 40% acetonitrile.

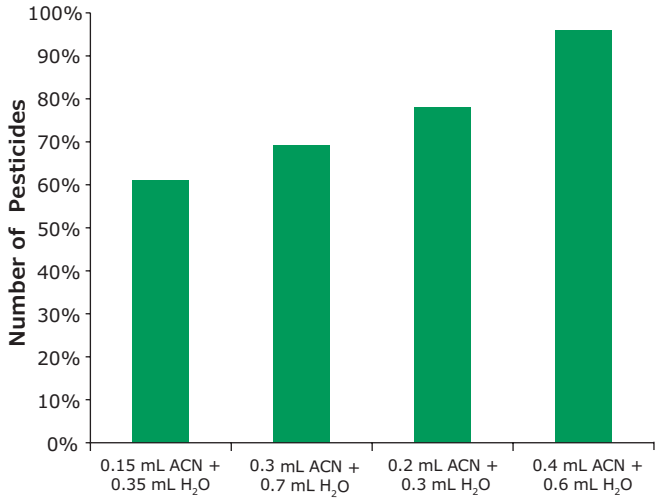
Even with these percentages of acetonitrile, sediment was observed in the extracts cleaned with PSA/C18, while the Z-Sep cleaned extracts were clear. Almonds were the more difficult matrix to analyze. Out of 113 pesticides analyzed, at a spiking level of 50 µg/kg, 94 had recoveries ranging from 70 to 120%. At a spiking level of 10 µg/kg, 92 pesticides had recoveries in this range. From avocado, 107 of the 113 pesticides had recoveries of 70-120% at both 10 and 50 µg/kg spiking levels.

To find the abstract & link to this article, see references at **SigmaAldrich.com/zsep**

(a) Percentage of total number of evaluated pesticides with recoveries in the range of 70 to 120%, in avocado. Yellow bars: evaporation of 0.50 mL, reconstitution in 0.05 mL of ACN + 0.45 mL of H2O; Green bars: evaporation of 0.50 mL, reconstitution in 0.30 mL of ACN + 0.70 mL of H_2O.



(b) Percentage of total number of evaluated pesticides with recoveries in the range of 70 to 120% in almonds using Z-Sep sorbent. Comparison of different reconstitution methods after evaporation of 0.5 mL.



GC Analysis of Pesticide Residues in Hops on SLB®-5ms after Extraction and Cleanup using Supel™ QuE Verde

There are a variety of pesticides used in the commercial production of hops. Hops contain essential oils, acids, pigments, and other compounds that present potential interferences. A standard approach to cleanup of hop extracts is the use of a PSA/C18/GCB sorbent blend. PSA removes acidic interferences while C18 removes hydrophobic interferences. GCB removes pigment. The compounds most effectively removed by commonly used GCB have planar structures. Any targeted pesticides with planar structures can also be retained.

In this application, Supel™ QuE Verde is used for QuEChERS cleanup in the analysis of pesticide residues in hops.

Supel™ QuE Verde consists of PSA, Z-Sep+ and an improved GCB, which provides improved/superior recoveries of planar pesticides compared to traditional while still maintaining sufficient removal of the chlorophyll background.

Sample Preparation:	
Sample/ Matrix	**Ground hop pellets, 1 g, spiked at 50 ng/g**
Extraction Process	1 g of ground sample. Add 10 mL of water to sample. Allow to sit for 30 minutes. Add 10 mL of acetonitrile containing 1% acetic acid; (shake for 10 min at 2500 rpm on automated shaker; add contents of Supel™ QuE Citrate extraction tube (55227-U); shake for 1 minute; centrifuge at 5000 rpm for 5 min; remove supernatant)
Cleanup Process	Transfer 1 mL of supernatant to the Supel™ QuE Verde cleanup tube, 2 mL (55447-U); shake for 2 min; centrifuge at 5000 rpm for 5 min; remove supernatant; place supernatant in amber low adsorption vial for GC-MS/MS analysis

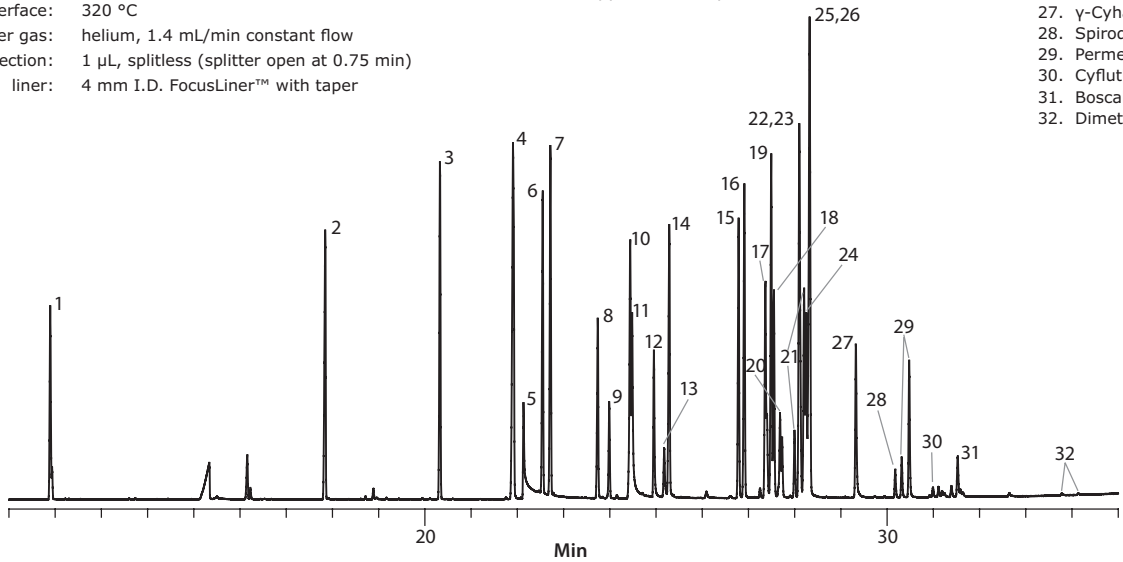
Results

Compound	% Rec	% RSD
Dichlorvos	68%	12%
Propoxur	86%	14%
Diazinon	101%	10%
Metalaxyl	83%	16%
Malathion	91%	13%
Chloropyrifos	97%	13%
Fipronil	89%	15%
Tetrachlorvinphos	82%	12%
Paclobutrazol	96%	10%
Myclobutanil	79%	7%
Pymetrozine	78%	7%
Trifloxystrobin	93%	9%
Quinoxyfen	64%	23%
Spiromesifens	91%	19%
Piperonyl butoxide	90%	10%
Methoxychlor	101%	18%
Permethrins	76%	8%
Boscalid	76%	24%
Cypermethrins	87%	14%
Pyraclostrobin	107%	17%
Dimethomorph	58%	15%

Analysis of a Standard Mix

column: SLB®-5ms, 30 m × 0.25 mm I.D. × 0.25 µm (28471-U)
oven: 50 °C (2 min), 8 °C/min to 320 °C (5 min)
inj. temp.: 250 °C
detector: MS/MS
MSD interface: 320 °C
carrier gas: helium, 1.4 mL/min constant flow
injection: 1 µL, splitless (splitter open at 0.75 min)
liner: 4 mm I.D. FocusLiner™ with taper

1. Dichlorvos
2. Propoxur
3. Diazinon
4. Metalaxyl
5. Propargite
6. Malathion
7. Chloropyrifos
8. Fipronil
9. Captan
10. Tetrachlorvinphos
11. Paclobutrazol
12. Imazalil
13. Pymetrozine
14. Myclobutanil
15. Trifloxystrobin
16. Quinoxyfen
17. Tebuconazole
18. Resmethrin isomers
19. Piperonyl butoxide
20. Spiromesifen isomers
21. Tetramethrin isomers
22. Phosmet
23. Bifenthrin
24. Fenoxycarb
25. Etoxazole
26. Methoxychlor
27. γ-Cyhalothrin
28. Spirodiclofen
29. Permethrin isomers
30. Cyfluthrin isomers
31. Boscalid
32. Dimethomorph isomers



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