

Solid Phase Extraction Products

Improve Sensitivity,
Increase Throughput
and Ensure Reliability



The life science business of Merck KGaA, Darmstadt, Germany operates as MilliporeSigma in the U.S. and Canada.

A Brief History of our Solid Phase Extraction (SPE) Products

SPE technology was first introduced by our company under the Supelclean™ and LiChrolut® brand names, with the introduction of our Visiprep™ Vacuum Manifold system shortly thereafter. With the focus on environmental, food/agrochemical and industrial analyses in the 1990s, we improved and extended the line further to include Supelclean™ ENVI™ SPE products. The late 1990s brought about the introduction of the Discovery® SPE line for pharmaceutical analysis.

Most recently, the emphasis for our Sample Prep R&D has been innovation. From the general “universal” polymeric SPE (i.e. Supel™-Select and LiChrolut® EN) to highly specialized products aimed at removing specific matrix interferences (i.e. HybridSPE®-Phospholipid and Supel™ QuE Z-Sep), our products enable chemists to quantitate their analytes of interest down to the lowest detection levels.



Supelclean™ and LiChrolut® columns

- Original pioneers of commercially available SPE Products
- Referenced in 100s of publications
- Developed, tested and quality controlled for environmental applications
- Also available in glass tubes and disk formats
- Unique chemistries such as ENVI-Carb™
- Documented applications in compliance to standardized EPA methods

Discovery® SPE

- Developed, tested and quality controlled for pharmaceutical and clinical applications
- Over 12 different phase chemistries ranging from mixed-mode SPE to polyamide adsorbents
- Available in 96-well and cartridge configurations
- Ultra-clean phases for highly sensitive analyses

An Era of Innovative SPE

- Supel™ Genie and LiChrospher® ADS Online SPE for high throughput and elimination of human error
- HybridSPE®-Phospholipid for quick and easy phospholipid and protein removal or phospholipid enrichment
- Supel™ QuE (dispersive SPE) for multi-residue pesticide analysis using the QuEChERS method
 - Z-Sep, Z-Sep/C18, Z-Sep+, and Verde sorbents for lipid and pigment removal
- Supel™-Select SPE polymeric SPE phases for extraction of a broad range of compounds from aqueous matrices.
- Supel™ Tox and Supelclean™ Ultra & EZ-POP NP, specialty phases for cleanup of mycotoxins, pesticides, and/or non-polar compounds in complex food matrices

Supelclean™ Specifications

| | |
|---------------------|--|
| Base Silica | Irregular, acid washed for ENVI |
| Mean Particle Size | 45 µm |
| Mean Pore Diam. | 60 Å |
| Tot. Pore Vol. | 0.8 cm ³ /g |
| Specific Surf. Area | 475 m ² /g |
| Endcapped | Yes (unless otherwise noted) |
| Frit | Polyethylene (PE), 20 µm porosity (unless otherwise noted) |

Discovery® Specifications

| | |
|---------------------|--|
| Base Silica | Irregular, acid washed |
| Mean Particle Size | 50 µm |
| Mean Pore Diam. | 70 Å |
| Tot. Pore Vol. | 0.9 cm ³ /g |
| Specific Surf. Area | 480 m ² /g |
| Endcapped | Yes (unless otherwise noted) |
| Frit | Polyethylene (PE), 20 µm porosity (unless otherwise noted) |

LiChrolut® Specifications

| | |
|---------------------|-----------------------------|
| Base Silica | Irregular, acid washed |
| Mean Particle Size | 40 – 63 µm |
| Mean Pore Diam. | 60 Å |
| Tot. Pore Vol. | 0.8 cm ³ /g |
| Specific Surf. Area | ~ 600 m ² /g |
| Encapped | No (unless otherwise noted) |
| Frit | Polyethylene (PE) |

The Importance of SPE

Solid phase extraction is a form of digital (on/off) chromatography designed to extract, partition and/or adsorb one or more components from a liquid phase (sample) onto stationary phase (sorbent or resin). Over the last twenty five years, SPE has become the most powerful technique available for rapid and selective sample preparation (prep) prior to analytical chromatography.

SPE extends a chromatographic system's lifetime and improves qualitative and quantitative analysis. Also, by changing an analyte of interest's original matrix environment to a simpler matrix more suitable for subsequent analysis, the demand placed on an analytical instrument is considerably lessened.

Figure 1. Urine sample without and with cleanup



For more applications and application details, visit SigmaAldrich.com/spe

Use SPE for Samples that:

- Require cleanup, trace enrichment/concentration or purification
- Contain particulate matter causing system clogging and high back-pressure
- Contain components that cause high background, misleading peaks and/or poor sensitivity
- Require sample matrix or solvent exchange

Benefits of SPE:

- Switch sample matrices to a form more compatible with chromatographic analyses
- Concentrate analytes for increased sensitivity
- Remove interferences to simplify chromatography and improve quantitation
- Protect the analytical column from contaminants

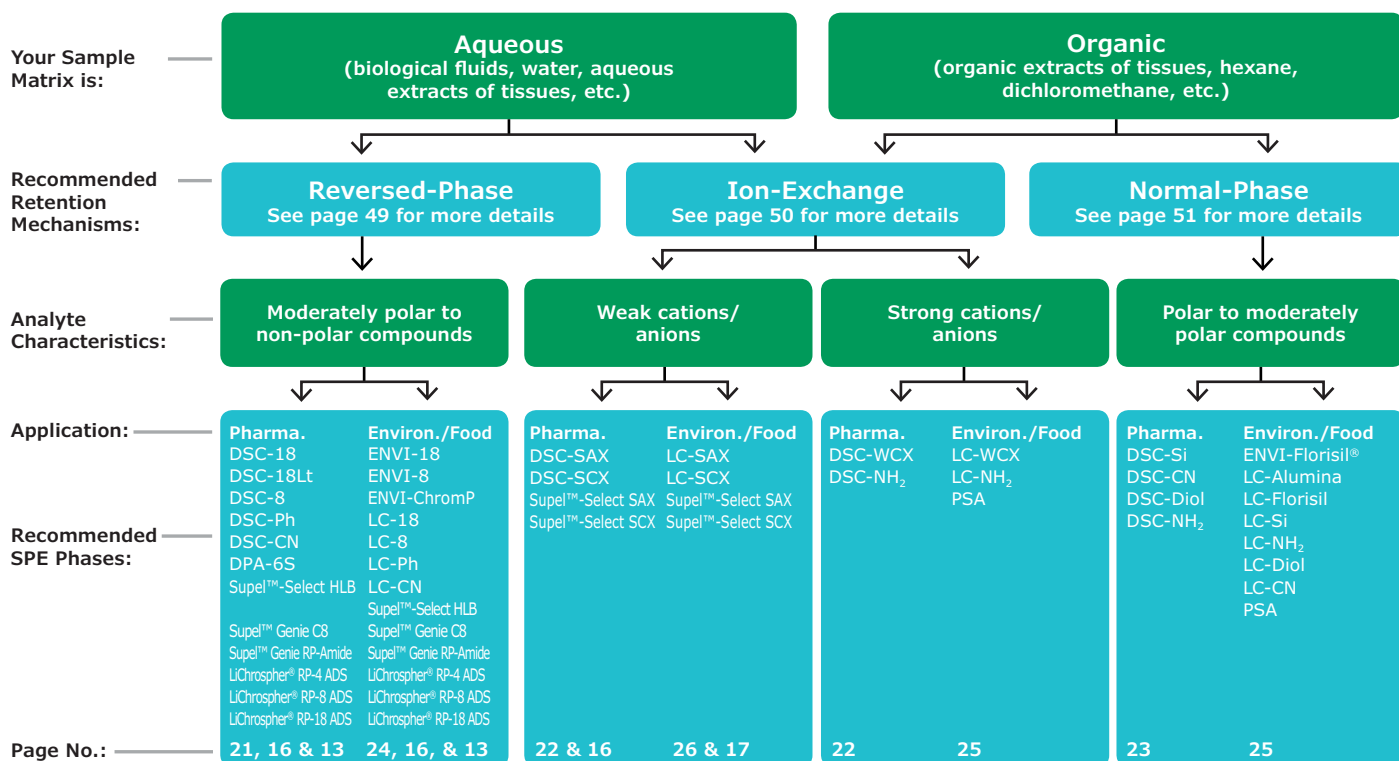
Common SPE Applications:

- Pharmaceutical compounds and metabolites in biological fluids
- Drugs of abuse in biological fluids
- Environmental pollutants in drinking and wastewater
- Pesticides, antibiotics or mycotoxins in food/agricultural matrices
- Desalting of proteins and peptides
- Fractionation of lipids
- Water and fat soluble vitamins

New and Featured Products

| Phase | Page | Description |
|--|------|--|
| HybridSPE®-Phospholipid | 8 | Combines the simplicity of protein precipitation with the selectivity of SPE for the targeted removal of proteins and phospholipids in biological samples. |
| Supel™ Genie and LiChrospher® ADS Online SPE | 11 | Hands free SPE done on the LC instrument to eliminate human error and increase throughput |
| Supel™-Select HLB, SAX, SCX | 16 | Hydrophilic polymer for extraction of a broad range of diverse analytes from aqueous samples. |
| EXtrelut® NT | 28 | Provides effective, emulsion-free solid-liquid extraction (SLE) using diatomaceous earth |
| Supelclean™ Ultra | 33 | Enhances recovery of pesticides from difficult, dry commodities (teas, spices, etc.) |
| Supel® QuE Z-Sep Sorbents | 35 | Enhance sample cleanup for complex matrices by removing more fat and color from sample extracts than traditional phases for QuEChERS methods. |
| Supel™ QuE Verde | 36 | Improves recoveries of planar compounds in green matrices. |
| Supel™ Tox | 39 | Removes interferences associated with mycotoxin analysis. |
| Supelclean™ EZ-POP NP | 41 | Removes oily matrix interferences for the analysis of lipophilic persistent organic pollutants (POPs) |

SPE Phase Selection Quick Look-Up Guide



Supelco SPE Specialty Phases

| Application | Field/ Application | Product | Page |
|---|-----------------------|--|---------|
| Phospholipid removal/enrichment | Ph | HybridSPE®-Phospholipid | 8 |
| Phospholipid removal in a pipette tip format | Ph | HybridSPE® DPX® Tips | 10 |
| Online SPE | Ph, G, E, F | Supel™ Genie and LiChrospher® ADS | 11 - 15 |
| Extraction of broad range of diverse analytes from aqueous samples | Ph, G, F | Supel™-Select HLB, SAX, SCX, and LiChrolut® EN | 16 - 18 |
| Molecularly Imprinted Polymer SPE | Ph, F, E | SupelMIP® SPE | 19 - 20 |
| Adsorption of polar compounds from aqueous or methanolic solution | G, E, Ph | Discovery® DPA-6S | 21 |
| Isolation of basic compounds from biological fluids | Ph, G | Discovery® DSC-MCAX | 22 |
| SPE filter discs (EPA 500 methods) | E | Supelclean™ ENVI-18 and -8 DSK SPE Disks | 24 |
| Desalting proteins/peptides and other macromolecules | B | Supelclean™ LC-4 (wide pore) | 24 |
| Removal or isolation of polar compounds from organic matrices | E | Dual Layer Florisil®/Na ₂ SO ₄ | 25 |
| Solid-liquid extraction (SLE) | Ph, F, E, G | EXTrelut® NT | 28 - 30 |
| Nitrosamines in water (EPA Method 521) | E | Supelclean™ Coconut Charcoal | 31 |
| Polar compounds in water | E | Supelclean™ ENVI-Carb™ Plus | 31 |
| PCBs from transformer/waste oils | E | Supelclean™ Sulfoxide | 31 |
| Pesticide residue analysis | F | Supelclean™ ENVI-Carb™ | 32 |
| Pesticide residue analysis | F | Multi-layer Supelclean™ SPE Products | 32 |
| Pesticide residue analysis | F | Supel™ Sphere Carbon/NH ₂ | 34 |
| Pesticide residue analysis from dry commodities (tea, spices, etc.) | F | Supelclean™ Ultra | 33 |
| Pesticide residue analysis - QuEChERS | F | Supel™ QuE Z-Sep, Z-Sep/C18, Z-Sep+, and Verde | 35 - 38 |
| Mycotoxin analysis | F | Supel™ Tox Cartridges | 39 - 40 |
| Non-polar POP analysis in edible oils | F | Supelclean™ EZ-POP NP | 41 |
| FAMES (cis/trans) analysis | F | Discovery® Ag-Ion | 42 |

Key: Ph = Pharmaceutical/Drugs; F = Food ; E = Environmental; B = Biological macromolecules; G = General

SPE Bed Weight Quick Look-Up Guide

Choosing the Right Bed Weight and Tube Size

General guidelines for choosing the appropriate SPE tube size and bed weight configuration are listed in this table. Optimal method parameters and hardware/bed weight dimensions should be determined during method optimization and troubleshooting.

| Bed Weight | Tube Volume | Minimum Elution Vol. | Bed Capacity* |
|------------|-------------|----------------------|---------------|
| 50-100 mg | 1 mL | 100-200 μ L | 2.5-10 mg |
| 500 mg | 3 mL | 1-3 mL | 25-100 mg |
| 0.5-1 g | 6 mL | 2-6 mL | 25-100 mg |
| 2 g | 12 mL | 10-20 mL | 0.1-0.2 g |
| 5 g | 20 mL | 20-40 mL | 1.25-2.5 g |
| 10 g | 60 mL | 40-100 mL | 0.5-1 g |

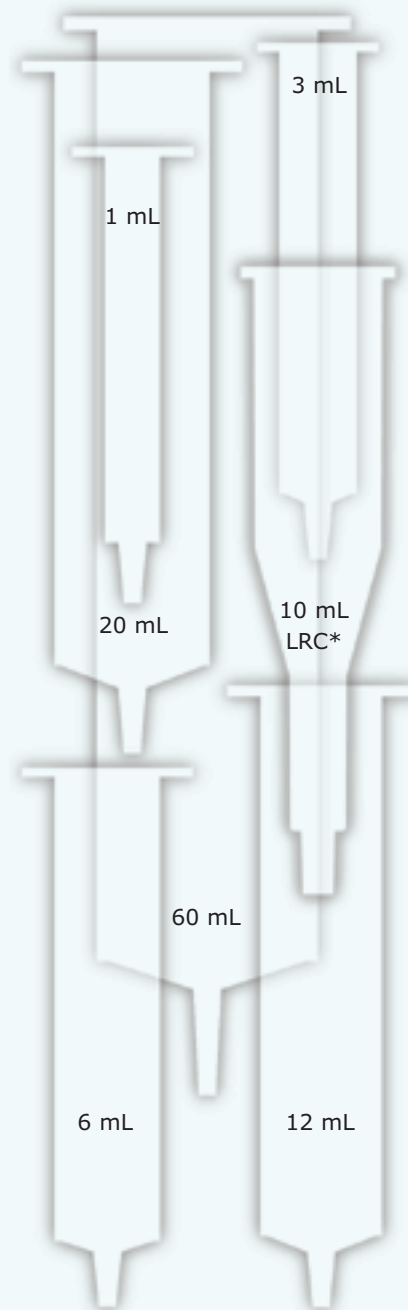
* This value depends on the analyte and sample matrix. As a rule of thumb, the bed capacity can be estimated with ~5% of the bed weight.

- Smaller tube dimensions (1 mL) contain smaller bed weights. Smaller bed weights allow for reduced elution volumes which can be beneficial for sensitive analyses, and when further processing is required (e.g., evaporation).
- 3 mL SPE tubes are the most common size dimension.
- 6 mL SPE tubes should be used when one or more steps in the SPE process require volumes greater than 3 mL. 6 mL tubes also contain larger bed weights (up to 1 g) which offers greater capacity, and can be beneficial when extracting difficult to retain compounds.
- 12, 20 and 60 mL tubes contain larger bed weights and head space volume which offer greater capacity. This allows researchers to use SPE as a purification or modified LPLC/Flash technique.
- The 10 mL LRC (large reservoir cartridges) are ideal for preparing larger sample volumes with smaller bed weights (25-100 mg). The packed section has the same diameter like a 1 mL tube.

FREE SPE MultiPaks for Method Development

SPE MultiPaks consist of an assortment of SPE phase chemistries and tube dimensions ideally suited for method development. The mix of phase chemistries available in these MultiPaks allows you to screen for optimal retention and selectivity required to achieve your sample prep objectives.

Figure 2. Most common SPE hardware: Polypropylene SPE tubes with PE Frit



* LRC: Large Reservoir Column

SPE Tubes and Specialty Hardware Quick Look-Up Guide

Additional Tubes and SPE Configurations

Glass SPE Tubes with PTFE and SS Frits (pg. 43)



Common in environmental analysis to reduce leachables from PP hardware and PE frits

Reversible SPE Tubes (pg. 31 and 43)



Reverse SPE tubes prior to elution to minimize elution volume for strongly retained compounds

SPE Disks (pg. 24: ENVI™-8 and ENVI™-18 DSK)



Allows for faster flow rates for processing large volume samples.

Discovery® SPE 96-Well Plates (pg. 21 - 23)



For high throughput sample preparation

Supel™ QuE (Dispersive SPE) for QuEChERS (pg. 35-38)



Salt and sorbent vials for dispersive SPE

Custom Capabilities

Supelco offers custom manufacturing services so you can optimize your sample processing procedure to the parameters dictated by your sample prep objectives. If there is a certain permutation of phase chemistry, bed weight and hardware configuration you require that is not listed within our standard product line, **contact your local Fisher Scientific sales representative.**

SPE Accessories Quick Look-Up Guide

SPE Manifolds

Visiprep™ DL and Standard Vacuum Manifold (pg. 44)



DL uses disposable liners that prevent cross-contamination

Visiprep™ 5-Port Flask Manifold (pg. 44)



Collects the SPE eluate in round flasks for easy rotary evaporation

Preppy™ Vacuum Manifold (pg. 45)



Most economical

PlatePrep Vacuum Manifold (pg. 47)



For 96-well SPE
Useful for stacking SPE tubes

ENVI-Disk™ Holder (pg. 48)



Used with 47 mm SPE disks

Visi-1™ Single SPE Tube Processor (pg. 44)



For processing very few
SPE samples

SPE Manifold Accessories

Visiprep™ Large Volume Sampler (pg. 45)



For processing larger
sample volumes

Visidry™ Drying Attachment (pg. 45)



For drying SPE tubes or
evaporating SPE eluate

SPE Tube Adapters and Large Volume Reservoirs (pg. 42)



Useful for stacking SPE tubes or
processing SPE tubes via luer
syringe; increasing tube volume

SPE Elution Rack (pg. 45)



Simple racks for using SPE
under gravity flow

Trap Kit and Vacuum Gauge Bleed Valve (pg. 46)



Additional vacuum accessories

HybridSPE®-Phospholipid Technology

Simultaneous protein and phospholipid removal

HybridSPE®-Phospholipid (HybridSPE®-PL) technology combines the simplicity of protein precipitation with the selectivity of solid phase extraction (SPE) for the targeted removal of phospholipids in biological plasma/serum (**Figure 3**). The technology utilizes a zirconia-coated particle, and exhibits selective affinity towards phospholipids while remaining non-selective towards a range of basic, acidic and neutral compounds. The phospholipid retention mechanism is based on a selective Lewis acid-base interaction between the proprietary zirconia ions (functionally bonded to the HybridSPE® stationary phase) and the phosphate moiety present in all phospholipids (**Figure 4**).



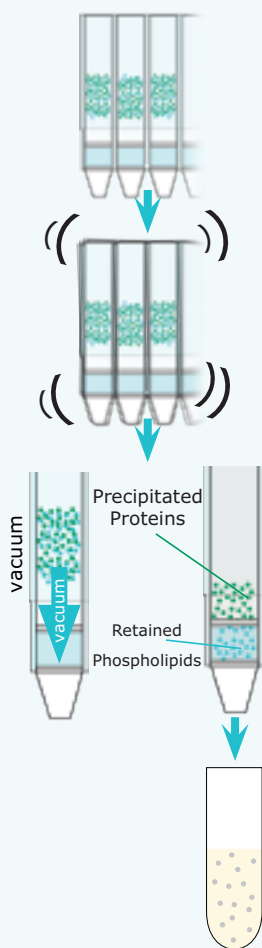
Figure 3. HybridSPE®-PL “In-well” Method

1. Precipitate Proteins by adding 100 μ L plasma or serum to the HybridSPE®-PL plate followed by 300 μ L 1% formic acid in acetonitrile. Add I.S. as necessary.

2. Mix by vortexing/shaking the HybridSPE®-PL plate or by aspirating/dispensing with 0.5-1 mL pipette tip (e.g., TOMTEC Quadra liquid handler).

3. Apply vacuum. The packed-bed filter/frit assembly acts as a depth filter for the concurrent physical removal of precipitated proteins and chemical removal of phospholipids. Small molecules (e.g., pharma compounds and metabolites) pass through unretained.

4. Resulting filtrate/eluante is free of proteins and phospholipids and ready for immediate LC-MS/MS analysis; or it can be evaporated and reconstituted as necessary prior to analysis.



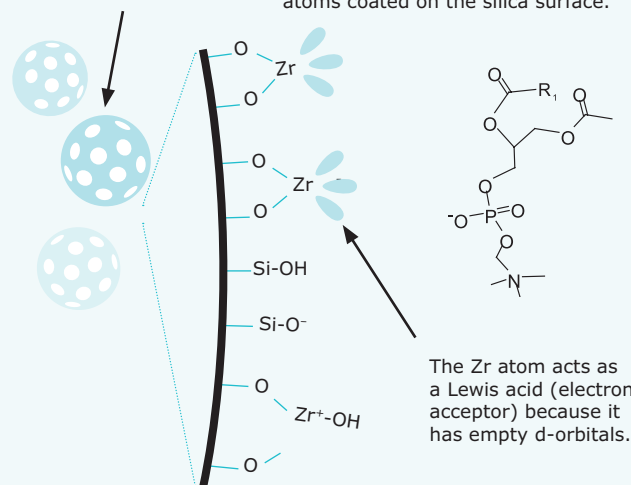
Features and Benefits

- Merges both protein precipitation and SPE
- Offers the simplicity of protein precipitation
- Selectively removes phospholipids via Lewis acid-base interactions
- 2-3 step generic procedure
- Typically >98% removal of phospholipids and precipitated proteins
- Minimal to no method development required
 - 96-well or individual cartridge format
 - Dispersive 96-well tip format (DPX®) and Online SPE formats (Supel™ Genie) offered on pages 11-12

Figure 4. Lewis Acid-Base Interactions Between HybridSPE® Zirconia atoms and Phospholipids

Proprietary HybridSPE® Zirconia Coated Silica

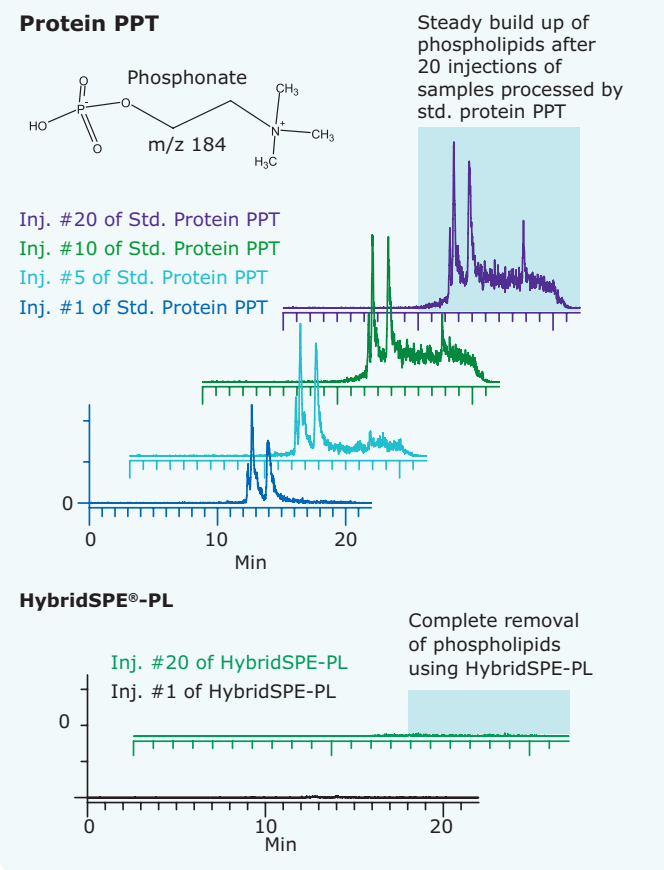
The phosphate moiety of phospholipids is a strong Lewis base (electron donor) that interacts with Zr atoms coated on the silica surface.



LC Accumulation of Phospholipids

With advances in LC-MS technology, many analysts are decreasing LC run time by incorporating ballistic gradients and sub-2 μm HPLC column particles. When coupled with standard protein precipitation (e.g., plasma:acetonitrile, 1:3 v/v), ballistic gradients are often inadequate at purging the column of phospholipids. As a result, phospholipids can build on the column (**Figure 5**), potentially change LC retention & selectivity, and elute uncontrollably downstream in an injection run sequence causing unpredictable ion-suppression effects and poor reproducibility. **Figure 5** compares a series of reversed-phase gradient LC-MS injections after standard protein PPT with HybridSPE®-PL in which m/z 184 (phosphonate moiety of phospholipids) is monitored. Unlike traditional protein PPT techniques that use centrifugation or simple filtration to remove precipitated proteins, HybridSPE®-PL 96-well plates contain a series of filters that allow users to concurrently remove proteins and phospholipids reducing LC column back pressure buildup commonly observed with standard PPT only, in particular for sub-2 μm HPLC columns that are more prone to clogging than larger particle size columns (2.7 - 5.0 μm) (**Figure 5**).

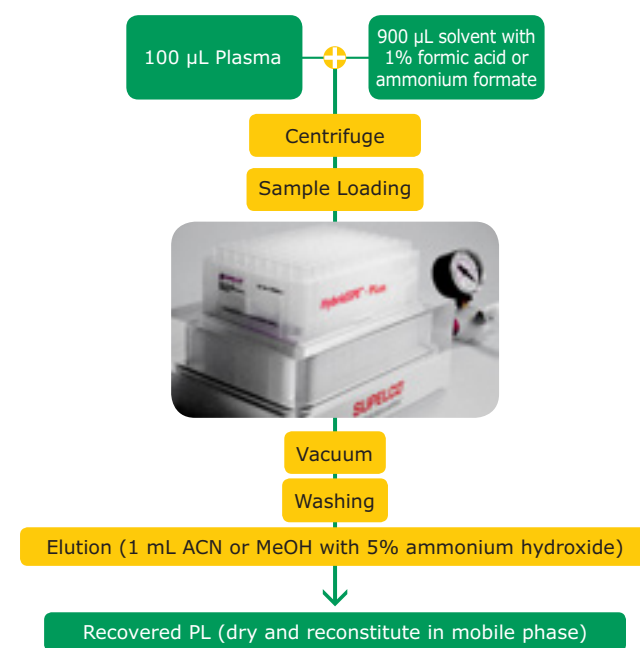
Figure 5. Gradient RP LC-MS of Blank Plasma Samples Prepared by Standard Protein PPT vs. HybridSPE®-PL



Phospholipid Enrichment Using HybridSPE®-Phospholipid Technology

Although HybridSPE®-Phospholipid is typically used to remove phospholipid interferences in biological samples, the same Lewis acid-base interactions that selectively remove phospholipids can also be used to recover phospholipids for analysis and phospholipid profiling. Phospholipids retained on the sorbent can be easily eluted with a strong basic solution, such as ammonium hydroxide. The bind and elute process of phospholipid enrichment is demonstrated in the flow chart below.

Figure 6. Experimental flow chart of the recovery of phospholipids from rabbit plasma



| Description | Qty. | Mfg. Cat. No. |
|---|------|---------------|
| Well Plates | | |
| HybridSPE®-PLus 96-well Plate, 50 mg/well | 1 | 575659-U |
| | 20 | 575673-U |
| HybridSPE®-PL, Small Vol. 96-well Plate, 15 mg/well | 1 | 52794-U |
| | 20 | 52798-U |
| HybridSPE®-PLus 96-Well Plate Essentials Kit (contains: 96-well Plate, 50 mg/well, 1 cap mat, sealing film, and collection plate) | 1 | 52818-U |
| SPE Cartridges | | |
| HybridSPE®-PL Ultra Cartridge, 30 mg/1 mL | 100 | 55269-U |
| HybridSPE®-PL Cartridge, 30 mg/1 mL | 100 | 55261-U |
| | 200 | 55276-U |
| HybridSPE®-PL Cartridge, 500 mg/6 mL | 30 | 55267-U |
| Plate Accessories | | |
| Round Well Cap Mat, Pierceable for HybridSPE®-PLus | 50 | 575680-U |
| 96 Round/Deep Well Collection Plate, PP for HybridSPE®-PLus | 60 | 2717266 |
| 96 Well-Plate Pre-cut Sealing Films | 100 | 2721581 |
| PlatePrep Vacuum Manifold | 1 | 57192-U |
| 96-well Protein Precipitation Filter Plate (for offline protein precipitation) | 1 | 55263-U |

Automated SPE with HybridSPE® DPX® Tips

Extraction in Seconds

DPX® stands for Dispersive Pipette Extraction. HybridSPE® DPX® Tips are pipette tips that incorporate loosely contained HybridSPE® sorbent material that is mixed with the sample solution when aspirated to accomplish solid phase extraction. HybridSPE® technology is a simple and generic sample prep platform designed for the gross level removal of endogenous phospholipid interferences from biological plasma and serum prior to LC-MS or LC-MS/MS analysis (see page 8).

In this simple technique, biological plasma or serum is first subjected to protein precipitation via the addition and mixing of acidified acetonitrile. Precipitated proteins are then removed by centrifugation and the resulting supernatant is extracted using the HybridSPE® DPX® tip which acts as a chemical filter that specifically targets the removal of endogenous sample phospholipids.

The phospholipid retention mechanism is based on a highly selective Lewis acid-base interaction between the proprietary zirconia ions functionally bonded to the HybridSPE® stationary phase and the phosphate moiety consistent with all phospholipids. The resulting eluent is ready for immediate LC-MS or LC-MS/MS analysis.

What size tips do I need?

| HybridSPE®-PL Sample and PPT Agent Guidelines | | |
|---|------------|------------|
| | 30 mg tips | 50 mg tips |
| Plasma/serum | 30-100 µL | 100-300 µL |
| Precipitating agent | 90-300 µL | 300-900 µL |

Figure 7. HybridSPE® DPX® Tips



The unique mixing technique employed provides numerous advantages:

- Minimal elution solvent volumes
- Rapid extraction times (less than 3 min. per sample/wellplate)
- High extraction efficiencies
- Easy to perform extractions
- Lower costs
- Higher throughput
- Minimal training required
- Environmentally friendly

| Description | Qty. | Mfg. Cat. No. |
|---|------|---------------|
| HybridSPE® DPX® tip, 30mg, Tecan® 200 µL | 96 | 52973-U |
| HybridSPE® DPX® tip, 50mg, Tecan® 1 mL | 96 | 52974-U |
| HybridSPE® DPX® tip, 30mg, Hamilton® 300 µL | 96 | 52977-U |
| HybridSPE® DPX® tip, 50mg, Hamilton® 1 mL | 96 | 52978-U |
| HybridSPE® DPX® tip, 30mg, Integra 300 µL | 96 | 52979-U |
| HybridSPE® DPX® tip, 50mg, Integra 1250 µL | 96 | 52980-U |
| HybridSPE® DPX® tip, 30mg, Universal 1mL | 96 | 52981-U |
| HybridSPE® DPX® tip, 50mg, Universal 1mL | 96 | 52982-U |

Go Hands Free with Online SPE

Supel™ Genie Online SPE Cartridges

Supel™ Genie Online SPE cartridges offer a sample preparation solution for a seamless workflow from start to finish performed entirely “online” using the LC instrument. Samples are directly injected onto the SPE cartridge located on the LC instrument for a simple and efficient hands-free solution.

How will Online SPE help you?

- Hands-free workflow
- Elimination of human error
- Decreased cost per sample
- Automation results in rapid throughput with greater reproducibility
- Clean samples leading to
 - Greater column life
 - Less instrument downtime
 - More accurate and reproducible data

We currently offer 3 phase chemistries:

- **HybridSPE®** - for complete removal of phospholipids (a leading cause of matrix effects) from biological samples (see previous pages for mechanisms)
- **C8** - for reversed-phase extraction of hydrophobic or nonpolar to moderately polar compounds
- **RP-Amide** - for reversed-phase extraction of nonpolar to polar compounds, compared to pure alkyl phases offers improved retention & performance for polar analytes, especially those that can interact via hydrogen bonding

Figure 8. Supel™ Genie HybridSPE® online starter kit (55324-U)



Figure 9. Supel™ Genie C8 online SPE cartridges, 2 pack (55512-U)



Supel™ Genie Online SPE Cartridges

HybridSPE® phase offers complete phospholipid removal from biological samples:

Figure 10. Phospholipids in Plasma Sample without Supel™ Genie HybridSPE® Online Cartridge (1st injection)

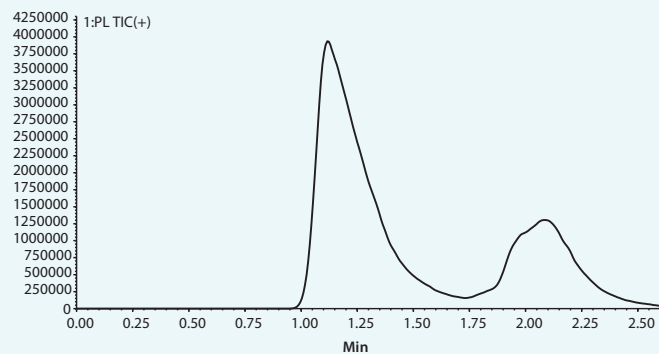
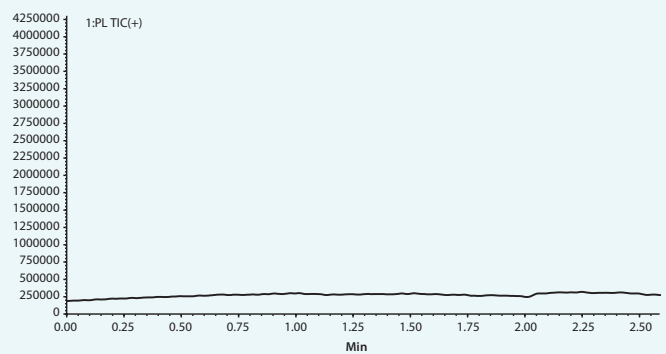


Figure 11. Phospholipids in Plasma Sample with Supel™ Genie HybridSPE® Online Cartridge (120th injection)



Check out our other applications at SigmaAldrich.com/onlinespe

Starter Kits come with reusable hardware that will fit any Supel™ Genie cartridge, as well as one cartridge of the selected phase chemistry. Additional cartridge packs include cartridges only.

HybridSPE® Products

| Description | Mfg. Cat. No. |
|--|---------------|
| Supel™ Genie HybridSPE® Online Starter Kit | 55324-U |
| Supel™ Genie HybridSPE® Online SPE Cartridge, pk. of 2 | 55326-U |
| Supel™ Genie HybridSPE® Online SPE Cartridge, pk. of 6 | 55327-U |

RP-Amide & C8 Products

| Description | Qty. | Mfg. Cat. No. |
|--|------|---------------|
| Supel™ Genie RP-Amide Online Starter Kit | | 55516-U |
| Supel™ Genie RP-Amide Online SPE Cartridge, pk. of 2 | | 55519-U |
| Supel™ Genie RP-Amide Online SPE Cartridge, pk. of 6 | | 55522-U |
| Supel™ Genie C8 Online Starter Kit | | 55274-U |
| Supel™ Genie C8 Online SPE Cartridge, pk. of 2 | | 55512-U |
| Supel™ Genie C8 Online SPE Cartridge, pk. of 6 | | 55515-U |



Need help choosing? Want more information on initial setup?

For more information or to order, [contact your local Fisher Scientific sales representative.](#)

LiChrospher® ADS Online Sample Prep Cartridges

LiChrospher® ADS allows the direct extraction and enrichment of hydrophobic, low molecular weight analytes from untreated samples such as hemolyzed blood, plasma, serum, milk, salivary fluid, fermentation broth, supernatants of cell cultures and tissue as well as food homogenates.

LiChrospher® ADS sorbents belong to the family of restricted access materials (RAM) with two chemically different surfaces, a hydrophilic external particle surface and a hydrophobic inner surface. Extraction and fractionation is based on the simultaneous performance of two chromatographic processes: reversed phase/ion-pair chromatography and size exclusion chromatography.

Specifications of LiChrospher® ADS

| | | |
|-------------------------------|--|--------------------------------|
| Sorbent characteristic | Spherical silica gel particles with two chemical different surface modifications | |
| Surface modifications | 1. Exterior surface | DIOL modification |
| | 2. Interior surface (surface of pores) | C-4, C-8, or C-18 modification |
| ADS | Alkyl-DIOL-Silica | |
| Particle size | 25 µm | |
| Pore diameter | 60 Å (6 nm) | |
| Stability | pH 2-7.5 | |

Benefits of LiChrospher® ADS at a glance

- Saves money and time: The high amount of analysis cycles, the direct injection of untreated biological fluids, and the fully automated system, extends column lifetime as well as saving time significantly
- Improved precision, accuracy, and sensitivity
- Quantitative elimination of protein matrix
- On-column enrichment of analytes



Figure 12. LiChrospher® ADS for direct on-line sample preparation

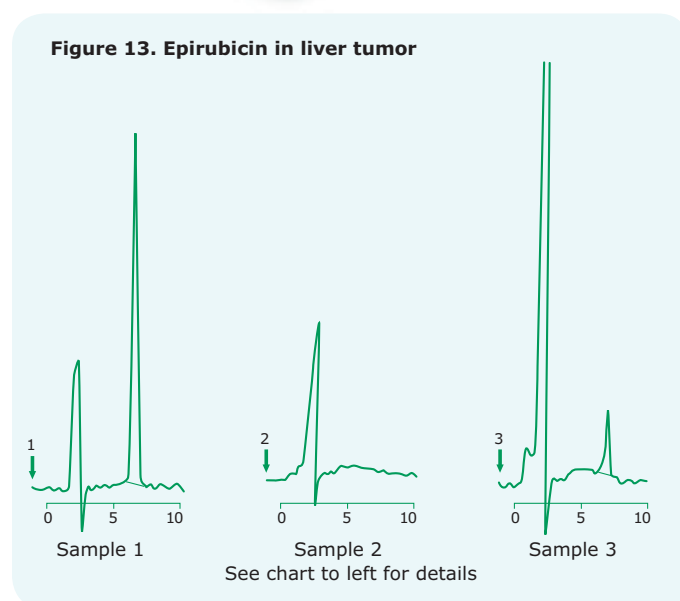
Applications of LiChrospher® ADS

Epirubicin in liver tumor

| | | |
|--------------------------|---|--------|
| Precolumn | LiChrospher® RP-4 ADS, 20 x 4 mm I.D. | |
| Analytical column | LiChrospher® 60 RP-select B, 250 x 4 mm I.D. | |
| Flow rate | 1 mL/min | |
| Loading | 95 % water, 5 % methanol | 10 min |
| Transfer | 30 % acetonitrile, 70 % water (0.1 % TEA, pH 2.0 with TCA) | 5 min |
| Separation | 30 % acetonitrile, 70 % water (0.1 % TEA, pH 2.0 with TCA) | 10 min |
| Detection | Fluorescence Ex 445 nm, Em 560 nm | |

Sample (50 µL)

1. Standard: 4'-Epirubicin-HCl, 31 mg/mL
2. Supernatant of liver homogenate (protein), 207 mg/mL
3. Supernatant of liver tumor homogenate (protein) after tumor chemoembolization with Lipiodol/4'-Epirubicin-emulsion, 1.34 mg/mL



LiChrospher® ADS Online Sample Prep Cartridges

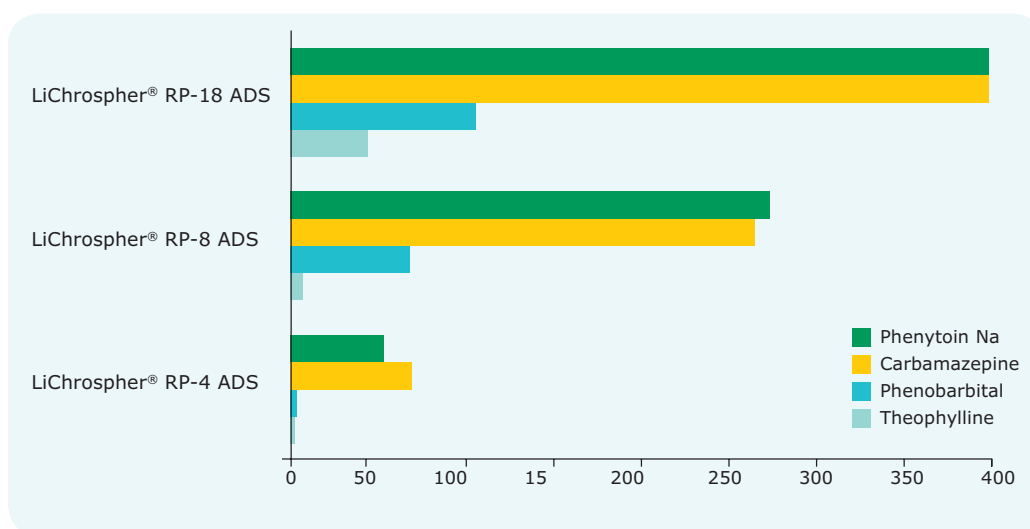
Choose the right column

The inner surface of the porous particles is exclusively covered with a hydrophobic dispersion phase (C4, C8, C18 alkyl chains). These adsorption centers are freely accessible for low molecular analytes. Owing to the classical reversed-phase chromatographic properties

of LiChrospher® RP ADS, these sorbents also can be used for ion-pair chromatography. This means that charged compounds can also be enriched and extracted by adding an appropriate ion-pair reagent (e.g. octanesulfonic acid) to the mobile phase.

Figure 14. Three types of LiChrospher® ADS cartridges are available showing different hydrophobicity, retention, and extraction properties for non-polar compounds

| LiChrospher® RP-4 ADS | LiChrospher® RP-8 ADS | LiChrospher® RP-18 ADS |
|-----------------------|-----------------------|------------------------|
| hydrophilic analytes | | hydrophobic analytes |



Selecting a LiChrospher® RP ADS cartridge with a lower hydrophobicity can be of advantage with respect to the sample transfer step, e.g. if the sample cleanup is performed using a LiChrospher® RP-8 ADS cartridge and the HPLC separation is performed on a RP-18 column, then it is possible to the lower amount of organic modifier for ADS elution, so that the transferred analyte fraction is enriched/re-focussed on the top of the analytical column.

LiChrospher® RP-4 ADS

| Description | Particle Size | Length | I.D. | Qty. | Mfg. Cat. No. |
|-------------------------------------|---------------|--------|------|---|---------------------|
| LiChrospher® RP-4 ADS | 25 µm | 25 mm | 2 mm | 1 piece | 1.50380.0001 |
| LiChrospher® RP-4 ADS | 25 µm | 25 mm | 2 mm | 3 pieces | 1.50381.0001 |
| LiChrospher® RP-4 ADS | 25 µm | 25 mm | 4 mm | 3 pieces | 1.50208.0001 |
| LiChrospher® RP-4 ADS cartridge set | 25 µm | 25 mm | 4 mm | 1 LiChrospher® RP-4 ADS 1 manu-CART® holder 25-4 | 1.50206.0001 |

LiChrospher® RP-8 ADS

| Description | Particle Size | Length | I.D. | Qty. | Mfg. Cat. No. |
|-------------------------------------|---------------|--------|------|---|---------------------|
| LiChrospher® RP-8 ADS | 25 µm | 25 mm | 2 mm | 1 piece | 1.50382.0001 |
| LiChrospher® RP-8 ADS | 25 µm | 25 mm | 4 mm | 3 pieces | 1.50209.0001 |
| LiChrospher® RP-8 ADS cartridge set | 25 µm | 25 mm | 4 mm | 1 LiChrospher® RP-8 ADS 1 manu-CART® holder 25-4 | 1.50207.0001 |

LiChrospher® RP-18 ADS

| Description | Particle Size | Length | I.D. | Qty. | Mfg. Cat. No. |
|--------------------------------------|---------------|--------|------|--|---------------|
| LiChrospher® RP-18 ADS | 25 µm | 25 mm | 2 mm | 1 piece | 1.50385.0001 |
| LiChrospher® RP-18 ADS | 25 µm | 25 mm | 2 mm | 3 pieces | 1.50386.0001 |
| LiChrospher® RP-18 ADS | 25 µm | 25 mm | 4 mm | 3 pieces | 1.50947.0001 |
| LiChrospher® RP-18 ADS cartridge set | 25 µm | 25 mm | 4 mm | 1 LiChrospher® RP-18 ADS 1 manu-CART® holder 25-4 | 1.50187.0001 |

LiChrospher® ADS cartridge kit and accessories

| Description | Particle Size | Length | I.D. | Qty. | Mfg. Cat. No. |
|--|---------------|--------|------|--|---------------|
| LiChrospher® ADS cartridge kit | 25 µm | 25 mm | 4 mm | 1 LiChrospher® RP-4 ADS 1 LiChrospher® RP-8 ADS 1 LiChrospher® RP-18 ADS 1 manu-CART® holder 25-4 | 1.50210.0001 |
| LiChrospher® ADS In-line filter (replacement pack) | 25 µm | - | - | 5 pieces | 1.51192.0001 |
| In-line filter holder | 25 µm | - | - | 1 piece | 1.51193.0001 |
| Filter insert In-line | 2 µm | - | - | 10 pieces | 1.51194.0001 |

LiChrospher® ADS bulk sorbents

| Description | Particle Size | Filling Amount | Packaging | Mfg. Cat. No. |
|-----------------------|---------------|----------------|----------------|---------------|
| LiChrospher® RP-4 ADS | 25 µm | 10 g | Plastic bottle | 1.50349.0010 |

For more information or to order, visit: fishersci.com/Supelco



Polymeric SPE

Supel™-Select

Features and Benefits

- Extract and recover a very broad range of compounds from aqueous samples
- Reduce ion-suppression
- Amenable to generic methodology
- Resistant to overdrying for greater reproducibility
- Low UV and MS extractables
- Stringent production and QC guidelines
- Greater capacity for smaller elution volumes

HLB and Ion-Exchange Phases for a Wide Range of Applications and pH Conditions

Supel™-Select SPE phases are ideal for the solid phase extraction of a broad range of compounds from aqueous samples. While reversed-phase interactions dominate retention on the Supel™-Select HLB, and the retention mechanisms of the Supel™-Select SAX and SCX are predominately based on ion-exchange, the hydrophilic modifications of the styrene-based polymer backbone allow also for retention and recovery of more polar compounds.

| | |
|----------------------|---|
| HLB Phase Chemistry: | Hydrophilic modified styrene polymer |
| SAX Phase Chemistry: | Quaternary amine functionalized hydrophilic modified styrene polymer; counter ion Cl ⁻ |
| SCX Phase Chemistry: | Sulfonic acid functionalized hydrophilic modified styrene polymer; counter ion H ⁺ |
| pH Compatibility: | 0-14 |
| Particle Size: | 50-70 μm |
| MS Suitable: | Yes |
| Surface Area: | 160-420 m ² /g |
| Pore Volume: | 0.8-1.2 mL/g |
| Pore Size: | 80-200 Å |

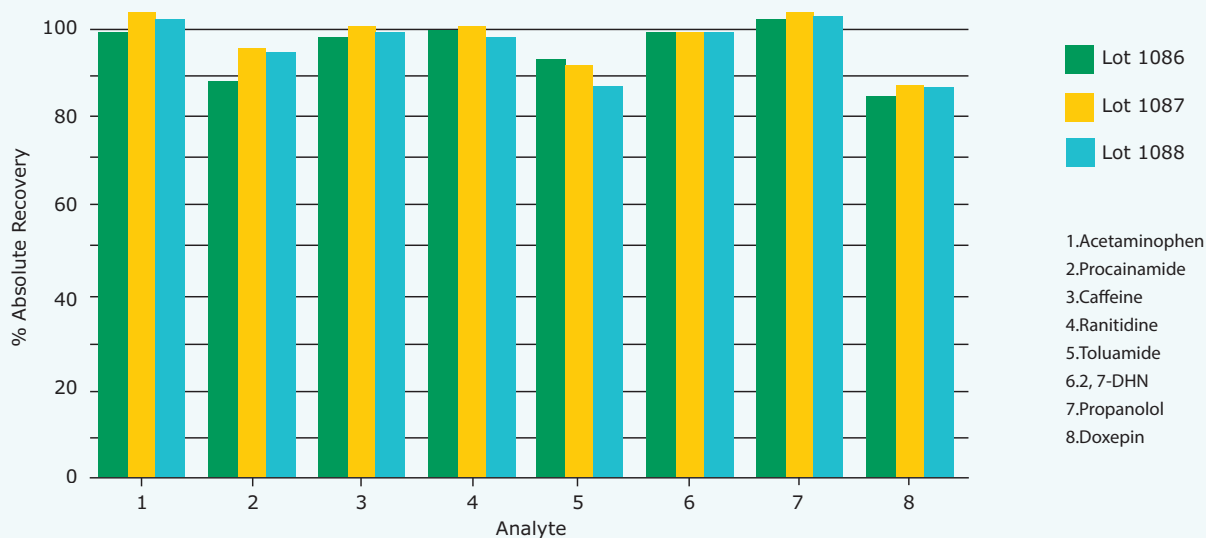
High and Reproducible Recoveries

The hydrophilic, lipophilic balanced Supel™-Select HLB SPE allows users to extract a broad range of compounds using a single sorbent and generic methodology. Analyte recovery was high across all the compounds tested, and results were highly reproducible across three production lots (see graphic below).

Figure 16. Supel™-Select Product Offering



Figure 15. Supel™-Select HLB Recoveries



Application: Isolation and LC-MS Characterization of Illicit Bath Salts in Urine

The analysis of bath salts from urine samples is demonstrated using polymeric SPE sample preparation, followed by hydrophilic interaction liquid chromatography (HILIC) analysis with TOF-MS detection. Supel™-Select SCX SPE is used for the processing and sample cleanup of the urine samples. The selective retention of the bath salts on the SCX cartridge is based upon the ion exchange mechanism between the anion functionality of the SCX and the basic functionality of the bath salts. The strong ionic interaction with the analytes enables high organic wash solvents to be used for displacement of the endogenous matrix, while maintaining retention of the analytes.

Elution of the bath salts is achieved with the addition of a basic organic solvent. This approach results in a very clean sample.

The figure below illustrates the monitored bath salt ions in a spiked urine sample after SPE cleanup (yellow), in a diluted spiked urine sample without cleanup (green) and in a urine blank after SCX cleanup (blue). Notice the chromatogram containing the bath salts in the spiked urine sample after SPE cleanup contains no interfering peaks. Therefore, the effectiveness of the SCX cleanup is demonstrated and the analysis is more robust and reliable.

Figure 17. LC-MS Analysis of Cathinones (Bath Salts) on the Ascentis® Express HILIC (Si) Column

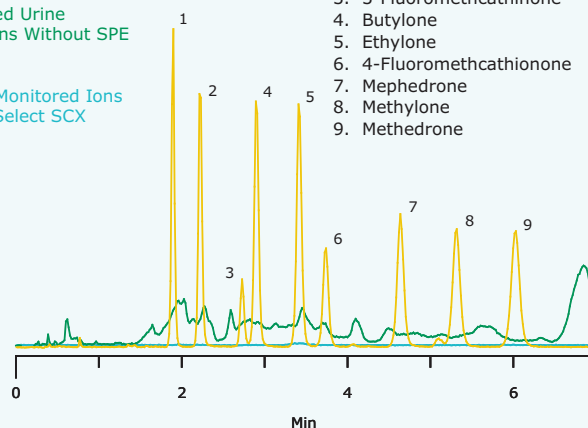
| | |
|------------------|--|
| sample/matrix: | 1 mL urine spiked to 100 ng/mL of bath salt mixture |
| SPE tube: | Supel™-Select SCX, 30 mg/1 mL (54240-U) |
| conditioning: | 1 mL 1% formic acid in acetonitrile, then 1 mL water |
| sample addition: | 1 mL spiked urine |
| washing: | 1 mL water, 1 mL 1% formic acid in acetonitrile, 1 mL water |
| elution: | 2 mL 10% ammonium hydroxide in acetonitrile |
| column: | Ascentis® Express HILIC (Si), 10 cm x 2.1 mm I.D., 2.7 µm (53939-U) |
| mobile phase: | (A) 5 mM ammonium formate acetonitrile; (B) 5 mM ammonium formate water; (98:2, A:B) |
| flow rate: | 0.6 mL/min |
| pressure: | 127 bar |
| column temp: | 35 °C |
| detector: | MS, ESI+, 100-1000 m/z |
| injection: | 1 µL |
| sample: | 200 ng/mL in acetonitrile |

Spiked Urine Sample
Monitored Ions After
Supel-Select SCX Cleanup

Diluted Spiked Urine
Monitored Ions Without SPE
Cleanup

Urine Blank Monitored Ions
After Supel-Select SCX
Cleanup

- 3,4-Methylenedioxypropylvalerone (MDPV)
- Buphenedrone
- 3-Fluoromethcathinone
- Butylone
- Ethylone
- 4-Fluoromethcathinone
- Mephedrone
- Methylone
- Methedrone



96-Well Plates

| Description | Qty. | Mfg. Cat. No. |
|--------------------------------------|------|--------------------------|
| Supel™-Select HLB 96-well SPE | | |
| 10 mg/ well | 1 | Inquire |
| 30 mg /well | 1 | 575661-U |
| 60 mg/ well | 1 | 575662-U |
| Supel™-Select SAX 96-well SPE | | |
| 10 mg/well | 1 | Inquire |
| 30 mg/well | 1 | 575660-U |
| 60 mg/well | 1 | 575663-U |
| Supel™-Select SCX 96-well SPE | | |
| 10 mg/well | 1 | Inquire |
| 30 mg/well | 1 | 575664-U |
| 60 mg/well | 1 | 575665-U |

SPE Tubes

| Description | Qty. | Mfg. Cat. No. |
|------------------------------|------|-------------------------|
| Supel™-Select HLB SPE | | |
| 30 mg/1 mL | 100 | 54181-U |
| 60 mg/3 mL | 50 | 54182-U |
| 200 mg/6 mL | 30 | 54183-U |
| 500 mg/12 mL | 20 | 54184-U |
| 1 g/20 mL | 20 | 54186-U |
| Supel™-Select SAX SPE | | |
| 30 mg/1 mL | 100 | 54231-U |
| 60 mg/3 mL | 50 | 54233-U |
| 200 mg/6 mL | 30 | 54235-U |
| 500 mg/12 mL | 20 | 54236-U |
| 1 g/20 mL | 20 | 54237-U |
| Supel™-Select SCX SPE | | |
| 30 mg/1 mL | 100 | 54240-U |
| 60 mg/3 mL | 50 | 54241-U |
| 200 mg/6 mL | 30 | 54242-U |
| 500 mg/12 mL | 20 | 54243-U |
| 1 g/20 mL | 20 | 54245-U |

LiChrolut® EN

High Capacity Polymeric Phase for Solid Phase Extraction

LiChrolut® EN resin was originally developed for environmental analysis applications for use with very polar organic compounds. In comparison to silica-based SPE phases, LiChrolut® EN resin has a ten-fold higher capacity. Thus, smaller amounts of sorbent suffice to provide reproducible extractions and high analyte recoveries.

Features and Benefits

- Use of common organic solvents, buffer solutions, acids and bases over the entire pH-range
- Solvent savings
- Time savings
- Increased sensitivity

LiChrolut® EN Specifications

| | |
|----------------------------|---|
| Sorbent type | Ethyl vinyl benzene divinyl benzene polymer |
| Particle shape | Irregular |
| Particle size distribution | 40 – 120 µm |
| Specific surface | 1,200 m ² /g (according to BET) |
| Pore volume | 0.75 mL/g |
| Stability | pH 1 – 13 |
| Capacity | 500 mg Caffeine/g sorbent (model substance for polar analytes) 500 mg Diisodecylphthalate DIDP/g sorbent (model substance for nonpolar analytes) |

LiChrolut® SPE Products

| Description | Qty. | Mfg. Cat. No. |
|----------------------------------|------|---------------|
| LiChrolut® EN (40 - 120 µm) | | |
| 200 mg/3 mL | 30 | 1.19693.0001 |
| 200 mg/3 mL* | 30 | 1.19870.0001 |
| 500 mg/6 mL | 30 | 1.19691.0001 |
| 200 mg/6 mL | 30 | 1.19941.0001 |
| LiChrolut® EN / RP-18 (top) | | |
| 100/200 mg/6 mL | 30 | 1.19912.0001 |
| LiChrolut® EN (40 - 120 µm) Bulk | | |
| 20 g | 1 | 1.19853.0020 |

*glass SPE tube



SupelMIP® SPE

Molecularly Imprinted Polymers

Features and Benefits

- Achieve lower detection limits through superior selectivity
- Reduce ion-suppression
- Minimal to no method development required, giving reduced sample prep time
- Stable at broad pH ranges and high temperatures

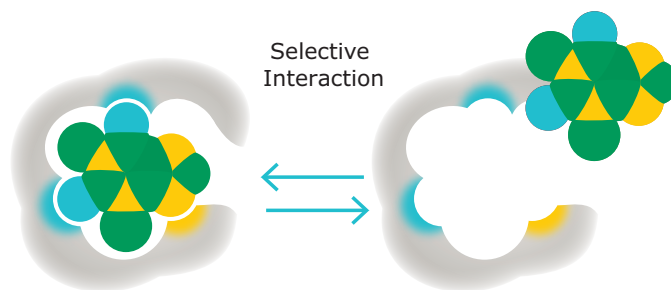
The SupelMIP® SPE line consists of highly cross-linked polymers that are engineered to extract a single analyte of interest or a class of structurally related analytes with an extremely high degree of selectivity. This is possible because selectivity is introduced during MIP synthesis in which a template molecule, designed to mimic the analyte, guides the formation of specific cavities or imprints that are sterically and chemically complementary to the analyte(s) of interest.

By careful design of the imprinting site, either by molecular modeling, experimental design or screening methods, the binding cavities can be engineered to offer multiple interaction points (ion-exchange, reversed-phase with polymer backbone, and hydrogen bonding) with the analyte(s) of interest. The MIP binding site is both chemically and sterically complementary to the analyte(s) of interest. This leads to a stronger interaction between the solid phase and the analyte(s). As a consequence, harsher wash conditions can be tolerated during SPE methodology, resulting in cleaner extracts. Because extraction selectivity is significantly improved, lower background is observed allowing analysts to achieve lower limits of detection.

SupelMIP® Phases and Methods available for:

- Aminoglycosides in animal tissue, cell culture, and honey
- β -agonists in tissue, urine, and wastewater• Clenbuterol in urine
- Bisphenol A (BPA) in broth or milk-based matrices
- Chloramphenicol in milk, plasma, honey, urine, and shrimp/prawns
- Fluoroquinolones in bovine kidney, honey, and milk
- Nitroimidazoles in milk, eggs, and other food matrices
- Non-steroidal anti-inflammatory drugs (NSAIDs) in wastewater and other sample matrices
- NNAL 4-(Methylnitrosamino)-1-(3-pyridyl)-1-butanol in urine
- TSNA (Tobacco Specific Nitrosamines) in urine and tobacco
- PAHs (polycyclic aromatic hydrocarbons) in edible oils
- Patulin in fruit matrices

Figure 18. SupelMIP® Selective Interaction

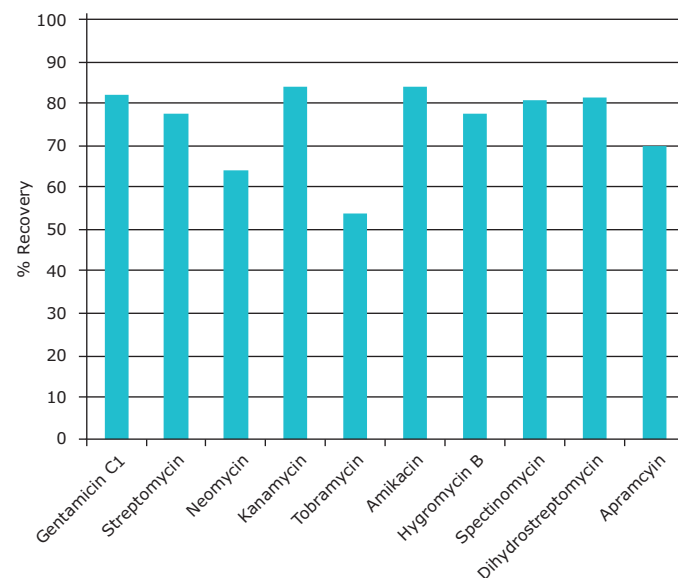


Application: Highly Selective Sample Preparation for the Analysis of Aminoglycoside Antibiotics in Pork Muscle

This study utilizes the unique extraction capabilities of MIPs to successfully quantitate ten aminoglycosides by LC-MS/MS at 100 ng/g (400 ng/g for neomycin) with recoveries $\geq 70\%$. The SPE cleanup procedure, using SupelMIP® SPE-Aminoglycosides, as well as the HPLC analysis, using an Ascentis® Express C18 HPLC column, is described in the condition section of **Figure 20**. Quantitation was performed using matrix-matched calibration standards, ranging from concentrations of 10 ng/mL to 1000 ng/mL.

Figure 20 depicts chromatograms of the analytes in pork muscle extracts. Recoveries for the 10 aminoglycosides are given in **Figure 19**. Most of the analyte recoveries were $\geq 70\%$, except for neomycin and tobramycin. Low recoveries for neomycin and tobramycin may be attributed to stronger binding of the analytes to the MIP sorbent due to the presence of several amino groups.

Figure 19. Aminoglycoside Recoveries in Pork Muscle Fortified at 100 ppb (400 ppb)

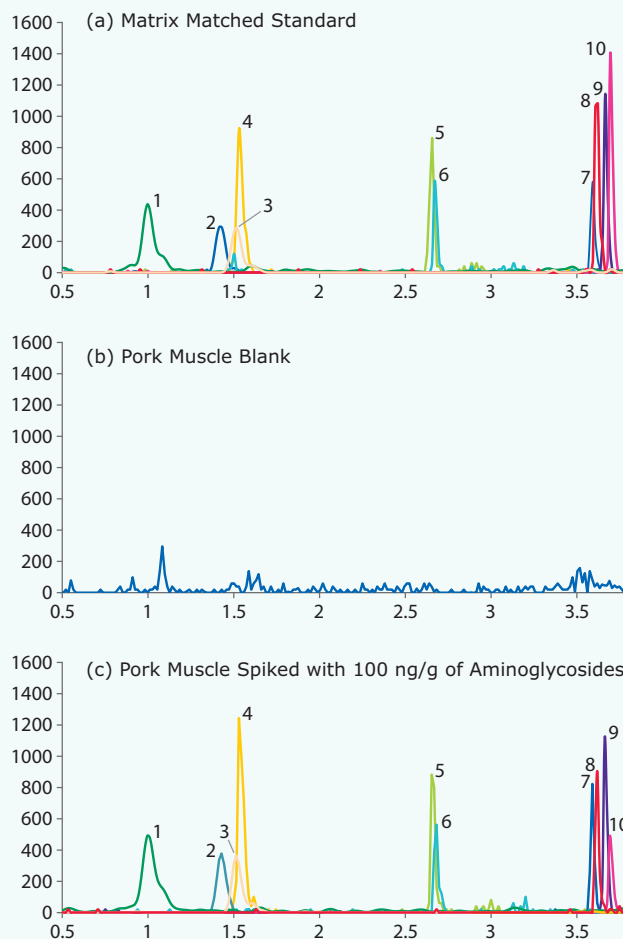


Molecularly Imprinted Polymers

Figure 20. LC-MS/MS Analysis of Aminoglycosides after SupelMIP® SPE Cleanup

sample/matrix: 3 mL of pork extract
 SPE tube/cartridge: SupelMIP® SPE – Aminoglycosides, 50 mg/3 mL (52777-U)
 conditioning: 1 mL of methanol, then 1 mL of 50 mM potassium phosphate in water (pH = 7.8)
 sample addition: 3 mL of pork extract
 washing: 3 mL of water, followed by drying with slight vacuum for 10 seconds
 washing: 1 mL of 50:50 dichloromethane:methanol (v/v), followed by drying with slight vacuum for 10 seconds
 elution: 1 mL of 1% formic acid containing 5 mM heptafluorobutyric acid (HFBA) in 80:20 water:acetonitrile (v/v)
 eluate post-treatment: thoroughly mix via vortex agitation, and transfer to polypropylene HPLC vials
 column: Ascentis® Express C18, 10 cm x 2.1 mm I.D., 2.7 µm (53823-U)
 mobile phase: (A) 5 mM heptafluorobutyric in water; (B) 5 mM heptafluorobutyric in acetonitrile
 gradient: 20 to 90% B in 3.0 min; held at 90% B for 1 min; 90 to 20% B in 0.1 min; held at 20% B for 5.9 min
 flow rate: 0.4 mL/min
 column temp.: 40 °C
 detector: MS/MS, ESI(+), MRM
 injection: 10 µL

| Peak ID | Precursor | Product |
|---------------------|-----------|---------|
| Spectinomycin | 351.1 | 333.1 |
| Hygromycin B | 528.1 | 177.1 |
| Streptomycin | 582.1 | 263.2 |
| Dihydrostreptomycin | 584.2 | 263.1 |
| Amikacin | 586.2 | 163.1 |
| Kanamycin | 485.2 | 163.1 |
| Apramycin | 540.2 | 217.1 |
| Tobramycin | 468.1 | 163.1 |
| Gentamicin C1 | 478.1 | 157.2 |
| Neomycin | 615.0 | 161.1 |



For additional information regarding this application, refer to an article from Supelco Reporter 32.2 available at SigmaAldrich.com/supelmip

| Description | 25 mg/3 mL pk 50 | 50 mg/3 mL pk 50 | 100 mg/3 mL pk 50 | 25 mg/10 mL (LRC) ¹ pk 50 | 96-well plates |
|---|---------------------|---------------------|----------------------------------|---|-------------------|
| Aminoglycosides | — | 52777-U | — | — | — |
| β-agonists (class selective) | 53225-U | — | — | 53202-U | — |
| Bisphenol A (BPA) | — | — | 52775-U, 54277-U ³ | — | — |
| Chloramphenicol | 53209-U | — | — | 53210-U | — |
| Clenbuterol | — | — | — | 53201-U | — |
| Fluoroquinolones | 53269-U | — | — | — | — |
| Nitroimidazoles | 52734-U | — | — | — | — |
| NSAIDs | 52769-U | — | — | — | — |
| NNAL (4-(methylnitrosamino)-1-(3-pyridyl)-1-butanol) | 53203-U | — | — | 53206-U | 53255-U |
| TSNAs (4 tobacco specific nitrosamines: NNK, NNN, NAB, NAT) | — | 53222-U | — | 53221-U ² | — |
| PAHs (Polycyclic Aromatic Hydrocarbons) | — | 52773-U | — | — | — |
| Patulin | — | — | 52776-U | — | — |

¹ LRC = large reservoir cartridge ² 50 mg/10 mL (LRC), pk 50 ³ 100 mg/6 mL, pk 50

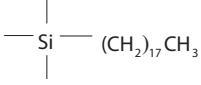
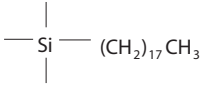
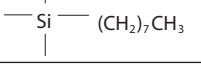
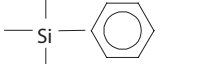
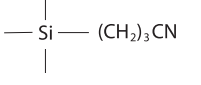
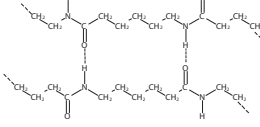
Discovery® SPE

Reversed-Phase

Discovery® reversed-phase SPE products are specifically developed, tested and quality controlled for pharmaceutical and clinical applications. Experience greater and more reproducible recoveries for the quick and effective extraction, isolation and concentration

of pharmaceuticals from biological fluids and other aqueous sample matrices.

For Discovery® silica specifications, see page 2. For general guidelines on reversed-phase SPE, see page 49.

| | |
|---|---|
| DSC-18  | <ul style="list-style-type: none"> • Polymerically bonded, octadecyl (18% C), endcapped • Higher 18% C loading for increased binding capacities and higher recoveries • The least selective phase: retains most organic analytes from aqueous matrices • Beneficial for extracting numerous analytes diverse in structure from the same sample |
| DSC-18Lt  | <ul style="list-style-type: none"> • Monomerically bonded, octadecyl (11% C), endcapped • Increased retention for moderately polar hydrophobic molecules • Used to elute very large hydrophobic molecules that are too strongly retained on DSC-18. Use this less retentive phase for the rapid release of hydrophobic compounds using weaker organic solvents at lower volumes |
| DSC-8  | <ul style="list-style-type: none"> • Monomerically bonded, octyl (9% C), endcapped; lower carbon content than DSC-18Lt • Used to elute very large hydrophobic molecules too strongly retained on DSC-18 or DSC-18Lt • Use this less retentive phase for the rapid release of hydrophobic molecules using weaker organic solvents at lower volumes |
| DSC-Ph  | <ul style="list-style-type: none"> • Monomerically bonded, phenyl (7% C), endcapped • Similar in polarity to DSC-8; however, electron dense aromatic ring offers some unique selectivity and retention |
| DSC-CN  | <ul style="list-style-type: none"> • Monomerically bonded, cyanopropyl (7% C), endcapped • Can behave as either reversed-phase or normal-phase • Ideal for very hydrophobic analytes that may be irreversibly retained on more hydrophobic sorbents such as DSC-18 • Less retentive than DSC-Si or DSC-Diol when used as normal phase (organic matrices such as hexane or oils) • Allows for the rapid release of very polar molecules irreversibly retained on very polar sorbents |
| DPA-6S  | <ul style="list-style-type: none"> • Polyamide Resin: Particle Size: 50-160 µm, Surf pH: 4.5-7.5, Density: 0.2-0.3 cm³/g, Water Content: <5% • Used to adsorb polar compounds (-OH groups, esp. phenolic compounds) from aqueous or methanolic solutions under the reversed-phase mechanism through strong hydrogen bonding between compound hydroxyl groups and amide groups of the resin • Useful for extracting tannins, chlorophyll, humic acid, pharmacologically active terpenoids, flavonoids, gallic acid, catechol A, protocatechuic acid and phloroglucinol • Also useful for extracting aromatic carboxylic acids, nitroaromatic compounds and irreversibly retains quinones |

Discovery® Reversed-Phase SPE Products

| Description | Qty. | DSC-18 | DSC-18Lt | DSC-8 | DSC-Ph | DSC-CN | DPA-6S |
|--------------------------------------|-------|----------|----------|---------|---------|---------|----------------------|
| Discovery® SPE Tubes | | | | | | | |
| 50 mg/1 mL | 108 | 52601-U | Custom | 52703-U | Custom | Custom | 52624-U |
| 100 mg/1 mL | 108 | 52602-U | 52611-U | 52707-U | Custom | 52694-U | Custom |
| 500 mg/3 mL | 54 | 52603-U | 52613-U | 52713-U | 52727-U | 52695-U | ¹ 52625-U |
| 500 mg/6 mL | 30 | 52604-U | 52615-U | 52714-U | 52728-U | 52696-U | ² 52626-U |
| 1 g/6 mL | 30 | 52606-U | 52616-U | 52716-U | Custom | 52697-U | ³ 52627-U |
| 2 g/12 mL | 20 | 52607-U | 52618-U | Custom | Custom | 52698-U | ⁴ 52629-U |
| 5 g/20 mL | 20 | 52608-U | Custom | Custom | Custom | Custom | ⁵ 52631-U |
| 10 g/60 mL | 16 | 52609-U | Custom | Custom | Custom | Custom | Custom |
| Discovery® SPE 96-Well Plates | | | | | | | |
| 100 mg/well | 1 | 575603-U | Custom | Custom | Custom | Custom | Custom |
| 50 mg/well | 1 | Custom | Custom | Custom | Custom | Custom | Custom |
| 25 mg/well | 1 | 575601-U | Custom | Custom | Custom | Custom | Custom |
| Bulk Packing | | | | | | | |
| | 100 g | 52600-U | | | | | ⁶ 52633-U |

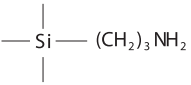
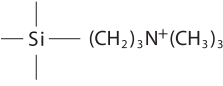

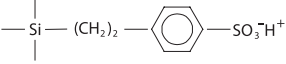
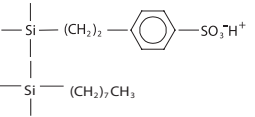
¹ 250 mg/3 mL, ² 250 mg/6 mL, ³ 500 mg/6 mL, ⁴ 1 g/12 mL, ⁵ 2 g/20 mL, ⁶ 50 g

Ion-Exchange and Mixed-Mode

Discovery® ion-exchange SPE products are specifically developed, tested and quality controlled for pharmaceutical and clinical applications. The Discovery® ion-exchange product line offers excellent selectivity towards charged molecular species enabling the user to extract, isolate, purify and concentrate charged ionizable pharmaceuticals (basic or acidic) from both polar and non-polar sample matrices.

Use mixed-mode SPE (e.g., Discovery® DSC-MCAX) for superior cleanup and selectivity when extracting basic pharmaceutical compounds from biological matrices such as plasma and urine.

For Discovery® silica specifications, see page 2. For general guidelines on ion-exchange and mixed-mode SPE, see page 50.

| | |
|--|--|
| DSC-NH₂  | <ul style="list-style-type: none"> • Polymerically bonded aminopropyl phase that is very polar in nature (hydrogen bonding) allowing for both normal-phase and ion-exchange applications • A weak anion exchanger with a pK_a of 9.8. At pH 7.8 or below, the functional groups are positively charged • Allows the rapid release of very strong anions such as sulfonic acids that may be retained irreversibly on SAX • Can be used in some reversed-phase applications (due to ethyl spacer); however, it is predominately used as an ion-exchanger or normal-phase sorbent due to its polar nature |
| DSC-SAX  | <ul style="list-style-type: none"> • A polymerically bonded quarternary amine that remains positively charged at all pH levels • Strong anion ion exchanger, commonly used when extracting weaker cations (e.g., carboxylic acids) that may not bind strongly enough to weaker anion exchangers • Selectivity can be modified by changing the counter ion with the appropriate buffer during conditioning • Counter ion is Cl⁻ |
| DSC-WCX  | <ul style="list-style-type: none"> • A polymerically bonded carboxy propyl phase with a K⁺ counter ion and a pK_a of 4.8 • Its weak cation exchange properties carries a negative charge at pH 6.8 or above • A pH of 2.8 or below neutralizes this phase for easier elution of strong cationic analytes that are neutralized only at extreme basic conditions • Typically used when dealing with very strong cationic (high pK_a) compounds that may be irreversibly retained on strong cation exchangers |
| DSC-SCX  | <ul style="list-style-type: none"> • A polymerically bonded, benzene sulfonic acid functional group with a H⁺ counter ion that is a strong cation exchanger due to its very low pK_a (<1.0) • Silica support allows for use with all common organic solvents (no shrinking/swelling) • Excellent capacity (0.8 meq/g) for cleaning up solution phase combinatorial chemistry reactions (removing target molecules from reaction by-products and excess reagents) • The presence of the benzene ring offers some mixed-mode capabilities (hydrophobic interactions) that should be considered when extracting cations from aqueous matrices |
| DSC-MCAX  | <ul style="list-style-type: none"> • Packed bed contains both octyl (C8) and benzene sulfonic acid (SCX) bondings. (H⁺ as counterion) • Developed for superior selectivity/sample cleanup when isolating basic compounds from biological fluids • Dual retention mechanisms broadens retention for a range of neutral, basic, acidic and zwitterionic compounds • Greater ion-exchange capacity for isolating polar basic and zwitterionic compounds • Can be used to fractionate basic/zwitterionic compounds from acidic and neutral compounds |

Discovery® Ion-Exchange SPE Products

| Description | Qty. | DSC-NH ₂ | DSC-SAX | DSC-WCX | DSC-SCX | DSC-MCAX |
|--------------------------------------|-------|---------------------|---------|---------|---------|-------------------------------|
| Discovery® SPE Tubes | | | | | | |
| 50 mg/1 mL | 108 | 52635-U | 52661-U | 52737-U | 52684-U | 52781-U |
| 100 mg/1 mL | 108 | 52636-U | 52662-U | 52739-U | 52685-U | 52782-U |
| 500 mg/3 mL | 54 | 52637-U | 52664-U | 52741-U | 52686-U | 52783-U ¹ |
| 500 mg/6 mL | 30 | 52638-U | 52665-U | Custom | 52688-U | 52784-U ² |
| 1 g/6 mL | 30 | 52640-U | 52666-U | 52743-U | 52689-U | 52788-U, 52786-U ³ |
| 2 g/12 mL | 20 | 52641-U | 52667-U | Custom | 52690-U | — |
| 5 g/20 mL | 20 | Custom | Custom | Custom | 52691-U | — |
| 10 g/60 mL | 16 | Custom | Custom | Custom | 52692-U | — |
| Discovery® SPE 96-Well Plates | | | | | | |
| 100 mg/well | 1 | 575615-U | Custom | Custom | Custom | Custom |
| 50 mg/well | 1 | Custom | Custom | Custom | Custom | Custom |
| 25 mg/well | 1 | Custom | Custom | Custom | Custom | Custom |
| Bulk Packing | | | | | | |
| | 100 g | 57212-U | 57214-U | 57228-U | 57221-U | — |

¹ 3 mL/100 mg, pk 54, ² 300 mg/3 mL, pk 54, ³ 300 mg/6 mL, pk 30

Normal-Phase

Discovery® normal-phase SPE products are specifically developed, tested and quality controlled for normal phase pharmaceutical applications and other modified flash techniques. The Discovery® normal phase product line enables you to quickly and effectively extract, isolate, purify and concentrate polar compounds from non-polar solutions. Its highly selective properties allow

the user to separate or remove structurally similar molecules through successive wash/elutions with increasingly polar solutions.

For Discovery® silica specifications, see page 2.
For general guidelines on normal-phase SPE, see page 51.

| | |
|---|---|
| DSC-Si $\begin{array}{c} \\ \text{—Si—OH} \\ \end{array}$ | <ul style="list-style-type: none"> • Unbonded acid washed silica sorbent ideal for normal-phase SPE and other modified flash techniques • Considered the most polar normal-phase sorbent available • Excellent capacity for purifying solution phase CombiChem reactions when removing target molecules from reaction by-products and excess reagents |
| DSC-Diol $\begin{array}{c} \\ \text{—Si—(CH}_2\text{)}_3\text{CH(OH)CH}_2 \\ \end{array}$ | <ul style="list-style-type: none"> • Polymerically bonded, 2,3-Dihydroxypropoxypropyl (7% C) • Polar sorbent most commonly used for normal-phase applications (polar extractions from non-polar matrices) • The sorbent's dihydroxy groups facilitate strong hydrogen bonding • Excellent selectivity when extracting structurally similar molecules |
| DSC-CN $\begin{array}{c} \\ \text{—Si—(CH}_2\text{)}_3\text{CN} \\ \end{array}$ | <ul style="list-style-type: none"> • Monomerically bonded, cyanopropyl (7% C), endcapped • Can behave as either reversed-phase or normal-phase • Ideal for very hydrophobic analytes that may be irreversibly retained on more hydrophobic sorbents such as DSC-18 • Less retentive than DSC-Si or DSC-Diol when used as normal-phase (organic matrices such as hexane or oils) • Allows for the rapid release of very polar molecules irreversibly retained on very polar sorbents |
| DSC-NH₂ $\begin{array}{c} \\ \text{—Si—(CH}_2\text{)}_3\text{NH}_2 \\ \end{array}$ | <ul style="list-style-type: none"> • Polymerically bonded, aminopropyl phase that is very polar in nature (hydrogen bonding) allowing for both normal-phase and ion-exchange applications • A weak anion exchanger with a pK_a of 9.8. At pH 7.8 or below, the functional groups are positively charged • Allows the rapid release of very strong anions such as sulfonic acids that may be retained irreversibly on SAX (a quarternary amine sorbent that is always positively charged) • Can be used in some reversed-phase applications (due to ethyl spacer); however, it is predominately used as an ion-exchanger or normal-phase sorbent due to its polar nature |

Discovery® Normal-Phase SPE Products

| Description | Qty. | DSC-CN | DSC-Si | DSC-Diol | DSC-NH ₂ |
|--------------------------------------|-------|---------|----------|----------|---------------------|
| Discovery® SPE Tubes | | | | | |
| 50 mg/1 mL | 108 | 52693-U | 52652-U | Custom | 52635-U |
| 100 mg/1 mL | 108 | 52694-U | 52653-U | 52748-U | 52636-U |
| 500 mg/3 mL | 54 | 52695-U | 52654-U | 52751-U | 52637-U |
| 500 mg/6 mL | 30 | 52696-U | 52655-U | 52752-U | 52638-U |
| 1 g/6 mL | 30 | 52697-U | 52656-U | 52753-U | 52640-U |
| 2 g/12 mL | 20 | Custom | 52657-U | Custom | 52641-U |
| 5 g/20 mL | 20 | 52699-U | 52658-U | Custom | 52642-U |
| 10 g/60 mL | 16 | 52700-U | 52659-U | Custom | 52644-U |
| Discovery® SPE 96-Well Plates | | | | | |
| 100 mg/well | 1 | Custom | Custom | Custom | 575615-U |
| 50 mg/well | 1 | Custom | 575608-U | Custom | Custom |
| 25 mg/well | 1 | Custom | Custom | Custom | Custom |
| Bulk Packing | | | | | |
| | 100 g | Custom | Custom | Custom | 57212-U |

Supelclean™ and Supelclean™ ENVI™ SPE

Reversed-Phase

The Supelclean™ SPE line represents one of our original brands. It is referenced in hundreds of journal publications and validated in methods such as EPA 500 series (drinking water) and SW-846 methods (solid waste).

For Supelclean™ silica specifications, see page 2. For general guidelines on reversed-phase SPE, see page 49.

| | |
|--|--|
| LC-18 | <ul style="list-style-type: none"> • Monomerically bonded, octadecyl (10% C), endcapped • For reversed-phase extraction of nonpolar to moderately polar compounds. • pH range 2-8 |
| LC-8 | <ul style="list-style-type: none"> • Monomerically bonded, octyl (7% C), endcapped |
| LC-4 (Wide Pore) | <ul style="list-style-type: none"> • Butyldimethyl, wide pore (500 Å), endcapped • Larger pore size to accommodate larger macromolecules (e.g., proteins and peptides) • Commonly used for desalting proteins and peptides in aqueous samples |
| LC-Ph | <ul style="list-style-type: none"> • Monomerically bonded, phenyl (5.5% C), endcapped |
| LC-CN | <ul style="list-style-type: none"> • Monomerically bonded, cyanopropyl (7% C), endcapped |
| Hisep™ | <ul style="list-style-type: none"> • Hydrophobic sites shielded by a hydrophilic surface for protein exclusion during sample load • Hydrophobicity similar to C8 |
| ENVI™-18 | <ul style="list-style-type: none"> • Polymerically bonded, octadecyl (17% C), endcapped • Excellent for cleaning, extracting and concentrating pollutants from aqueous environmental samples • Higher 17% C loading for increased binding capacities and higher recoveries • Higher carbon loading also offers greater resistance to extreme pH conditions • Typical applications include herbicides, fungicides, pesticides and other aqueous hazardous waste materials • Ideal for EPA 500 series including 525.1 and 508.1 |
| ENVI™-18 DSK and ENVI™-8 DSK SPE Disks | <ul style="list-style-type: none"> • The SPE membrane equivalents of ENVI™-18 and ENVI™-8 packed bed SPE sorbents • Porous glass fiber membranes embedded with C18 or C8 silica particles • Provides faster flow rates and exhibits less clogging than PTFE discs for the extraction of organic contaminants from drinking water • Typical applications include PAHs, PCBs, phthalates, semivolatile organics, paraquat and diquat, pesticides and herbicides • Ideal for EPA 500 series including 525.1 and 508.1 |
| ENVI™-8 | <ul style="list-style-type: none"> • Available in glass tubes with PTFE frits • High 14% C loading for increased binding capacities and higher recoveries • Higher carbon loading also offers greater resistance to extreme pH conditions • Excellent for cleaning, extracting and concentrating pollutants from aqueous environmental samples |
| ENVI™-Chrom P (polystyrene divinylbenzene) | <ul style="list-style-type: none"> • Styrene/divinylbenzene co-polymer resin: Particle Size: 80-160 µm; Spherical Shape; Pore Size: 110-175 Å; Surface Area: 900 m²/g • Highly crosslinked, neutral, specially cleaned styrene-divinylbenzene resin used to retain hydrophobic compounds with some hydrophilic functionality under the reversed-phase mechanism • Highly resistant to extreme pH conditions • Typical applications include aromatic and phenolic compounds from aqueous sample matrices • Used for priority pollutant phenols from aqueous samples |
| ENVI™-Carb and ENVI™-Carb II (graphitized carbon black) | <ul style="list-style-type: none"> • Surface Area: 120 m²/g, Particle Size: 100/400 mesh (ENVI™-Carb-II: 120/140 mesh) • Extreme affinity for organic polar and non-polar compounds from both non-polar 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 dispersion into solid phase pores • Independent investigators have found ENVI™-Carb extremely useful for the rapid sample preparation of over 200 pesticides from various matrices including ground water, fruits and vegetables (see publication T196900 on our web site) |

For available configurations and part numbers, please see page 26.

Ion-Exchange and Normal-Phase

The Supelclean™ SPE line represents one of the original brands to be introduced into the market place. It is referenced in hundreds of journal publications and validated in a variety of methods spanning environmental applications to the food and beverage industry. The Supelclean™ ENVI™ line was developed

and optimized for numerous environmental methods, including EPA 500 series (drinking water methods) and a number of SW-846 methods (solid waste).

For Supelclean™ silica specifications, see page 2. For general guidelines on ion-exchange and normal-phase SPE, see pages 50 and 51.

| | |
|--|---|
| LC-SAX | <ul style="list-style-type: none"> • A strong anion exchanger • Quarternary amine, Cl⁻ counter-ion |
| LC-SCX | <ul style="list-style-type: none"> • Aliphatic sulfonic acid, Na⁺ counter-ion, endcapped |
| LC-WCX | <ul style="list-style-type: none"> • Carboxylic acid, Na⁺ counter-ion |
| LC-NH ₂ | <ul style="list-style-type: none"> • Monomerically bonded, aminopropyl (5% C) |
| PSA | <ul style="list-style-type: none"> • Polymerically bonded, ethylenediamine-N-propyl phase that contains both primary and secondary amines with pK_a of 10.1 and 10.9 |
| ENVI™-Florisil® | <ul style="list-style-type: none"> • Magnesium silicate, mesh: 100/200, available with PTFE or stainless steel frits • Tested for US Environmental Protection Agency (EPA) Contract Laboratory Program (CLP) statement of work for pesticides • Highly polar material that strongly adsorbs polar compounds from non-polar matrices under normal-phase conditions • Typical applications include alcohols, aldehydes, amines, herbicides, pesticides, PCBs, ketones, nitro compounds, organic acids and phenols |
| Dual Layer Florisil®/Na ₂ SO ₄ | <ul style="list-style-type: none"> • Dual layer SPE Tube (available as glass or PP) that contains Na₂SO₄ (upper layer) and Florisil® (magnesium silicate; lower layer) separated and packed with PTFE frits • Florisil®, activated, size- 60/100 mesh (150-200 mm), Na₂SO₄ Purity- 99.99 %, Density- 2.68 g/mL • Excellent for removing/isolating polar compounds from organic matrices • Na₂SO₄ layer aids in removing aqueous sample residues that may hinder Florisil® performance and/or subsequent GC analysis • Suitable for the determination of the hydrocarbon oil index in water (surface, waste and sewage treatment plants) by GC/FID analysis according to European Standard EN ISO 9377-2:2000 (enclosed in the Extraction Kit for EN ISO 9377-2 Cat. No. 68172) • Use in conjunction with Visiprep™ Large Volume Sampler (Cat. No.57275, only suitable for the PP version with PE frits 54116-U) and Visiprep™ SPE Vacuum Manifolds for processing larger volume samples |
| LC-Florisil® | <ul style="list-style-type: none"> • Magnesium silicate, mesh: 100/120 |
| LC-Alumina A, N, and B | <ul style="list-style-type: none"> • Alumina-A for acidic pH (~5) • Alumina-N for neutral pH (~6.5) • Alumina-B for basic pH (~8.5) • Brockman Activation I for all Alumina SPE products, mesh: 60/325 |
| LC-CN | <ul style="list-style-type: none"> • Monomerically bonded, cyanopropyl (7% C), endcapped |
| LC-Si | <ul style="list-style-type: none"> • Silica gel |
| LC-Diol | <ul style="list-style-type: none"> • Monomerically bonded, Diol (7% C), endcapped |

For available configurations and part numbers, please see page 26.

All SPE tubes listed consist of polypropylene hardware and PE frits unless noted otherwise. Color coded

footnotes denote differences in hardware, package size or bed weight from the standard configuration.

| | Description | 0.1 g/1 mL pk 108 | 0.5 g/3 mL pk 54 | 0.5 g/6 mL pk 30 | 1 g/6 mL pk 30 | 2 g/12 mL pk 20 | 5 g/20 mL pk 20 | 10 g/60 mL pk 16 | 100 g bulk |
|------------------|---|----------------------|-----------------------|-----------------------|-------------------------|------------------------|--------------------|---------------------|------------------------|
| Reversed-Phase | ENVI™-18 | 57062 | 57063 | 57064 | 505706 | 57114 | 57137 | 57138 | 57219 |
| | ENVI™-18 DSK SPE Disks | | | ●54331-U ¹ | | | | | |
| | ENVI™-8 DSK SPE Disks | | | ●57171 ¹² | ●57170-U ¹³ | | | | |
| | ENVI™-8 DSK SPE Disks | | | ●57172 ¹² | | | | | |
| | LC-18 | 504270 | 57012 | 57054 | 505471 | 57117 | 57135-U | 57136 | 57202 |
| | ENVI™-8 | 57230-U | 57231 | 57232 | 57233 | | Custom | Custom | |
| | LC-8 | 504157 | ●Custom | ●57107 ¹ | | | | | 57201 |
| | ENVI™-Chrom P | 57143 | ●57224 ⁵ | 57052 | 57226 | | | | ●57217 ¹¹ |
| | ENVI™-Carb | 57109-U | ●57088 ⁵ | 57094 | | 57128 | Custom | 57130 | ●57210-U ¹¹ |
| | ENVI™-Carb C, mesh 80/100 | | | ●57092 ⁷ | | ●57127-U ¹⁰ | | | |
| LC-4 (Wide Pore) | | 57089 | | | ●57149 ¹⁰ | | | | |
| Hisep | | 57076-U | | | | | | | |
| LC-Ph | 504599 | 505269 | | | | | | | |
| LC-CN | 504386 | 57013 | 57056 | | | Custom | | | |
| LC-Diol | Custom | 57016 | | | | | | | |
| Normal-Phase | ENVI™-Florisil® | | ●57058 ² | ●57046 ³ | ●57053 ³ | | | | |
| | Dual Layer Florisil®/ Na ₂ SO ₄ | | | | ●54095-U ¹ | | | | |
| | Dual Layer Florisil®/ Na ₂ SO ₄ | | | | ●52582-U ^{1,9} | | | | |
| | Dual Layer Florisil®/ Na ₂ SO ₄ | | | | ●54116-U ^{2,9} | | | | |
| | LC-Florisil® | | | ●54333-U ¹ | 57057 | 57115 | 57131 | 57132 | 57209 |
| | LC-Florisil® | | | | ●54334-U ¹ | | | | |
| | LC-Alumina A | | ●57082-U ⁶ | | ●57083-U ⁸ | | | | 57026 |
| | LC-Alumina B | | ●57084 ⁶ | | ●57085 ⁸ | | | | 57207 |
| | LC-Alumina N | | ●57086 ⁶ | | ●57087 ⁸ | | | | 57028 |
| | LC-Si | 504041 | 505048 | 505374 | 57051 | 57116 | 57133 | 57134 | 57200 |
| LC-Si | | | | ●54335-U ¹ | | | | | |
| Ion Exch. | LC-NH ₂ | 504483 | 57014 | 54059-U | | | | | 57205 |
| | PSA | | ●52578-U ⁴ | 52579-U | | | | | 52738-U |
| | LC-SAX | 504815 | 57017 | | | | | | 57203 |
| | LC-SCX | 504920 | 57018 | | | | | | Custom |
| | LC-WCX | 505595 | 57061 | | | | | | |

Footnotes/Color Codes

●¹ glass SPE tubes, PTFE frits

●² PP SPE tubes, PTFE frits

●³ PP SPE tubes, stainless steel frits

●⁴ 0.2 g/3 mL, pk 54

●⁵ 0.25 g/3 mL, pk 54

●⁶ 1 g/3 mL, pk 54

●⁷ 0.25 g/6 mL

●⁸ 2 g/6 mL, pk 30

●⁹ 2 g/2 g/6 mL, pk 48

●¹⁰ 1 g/12 mL, pk 20

●¹¹ 50 g bulk

●¹² 47 mm diam. disks, pk 24

●¹³ 90 mm diam. disks, pk 12

Multi-Layer SPE

Developed to provide superior cleanup when conducting multi-residue pesticide analysis in food/agricultural matrices.

| Description | Qty. | Mfg. Cat. No. |
|------------------------------|------|----------------|
| ENVI™-Carb-II/PSA | | |
| 0.3 g/0.6 g/6 mL | 30 | 54058-U |
| 0.5 g/0.5 g/6 mL | 30 | 54067-U |
| 0.5 g/0.3 g/6 mL | 30 | 55119-U |
| 0.5 g/0.5 g/20 mL | 20 | 54217-U |
| ENVI™-Carb-II/SAX/PSA | | |
| 0.5 g/0.5 g/0.5 g/12 mL | 20 | 52574-U |
| SAX/PSA | | |
| 0.25 g/0.25 g/6 mL | 30 | 52576-U |
| 0.5 g/0.5 g/6 mL | 30 | 52577-U |

See also the new dual layer Supel™ Sphere products containing spherical materials on page 34.

| Description | Qty. | Mfg. Cat. No. |
|--|------|----------------|
| ENVI™-Carb/LC-NH₂ | | |
| 0.5 g/0.5 g/3 mL | 20 | 54332-U |
| 0.5 g/0.5 g/20 mL | 20 | 54216-U |
| 0.5 g/0.5 g/6 mL | 300 | 54024-U |
| 0.5 g/0.5 g/6 mL | 30 | 54035-U |
| ENVI™-Carb/NH₂/Silica | | |
| 0.5 g/0.4 g/0.6 g/12 mL | 20 | 54034-U |
| 0.5 g/0.4 g/0.6 g/20 mL | 20 | 54036-U |
| Dual Layer Florisil®/Na₂SO₄ | | |
| Glass tubes, PTFE frits, 2 g/2 g/6 mL | 48 | 52582-U |
| PP tube with PE frits 2 g/2 g/6 mL | 48 | 54116-U |

LiChrolut® SPE Products

Reverse Phase, Normal Phase & Ion Exchange

The LiChrolut® SPE line also represents one of our original brands. For LiChrolut® silica specifications, please refer to page 2. The table below contains

information about the typical applications for each LiChrolut® product. This selection guide will help you select the right product for your application needs.

| Application | LiChrolut® extraction column | Typical sample matrix | Typical sample substances | Typical elution solvent |
|---|-----------------------------------|--|---|---|
| Non-polar extraction | RP-18 RP-18e (endcapped) CN | Aqueous buffer solution | Aromatic ring systems, compounds with alkyl chains | Acetonitrile, methanol, ethyl acetate |
| Cation exchange extraction | SCX (strong) | Methanolic/aqueous buffer with low ionic strength; 2 pH units under pK value of the sample substance | Cations: amines, pyrimidines | Aqueous buffer of high ionic strength (0.1 mol/L); 2 pH units over pK value of the sample substance |
| Mixed mode extraction | TSC (Tox Screening Cation) | Body fluids (not for in vitro) | Cationic and neutral analytes | Chloroform-acetone, NH3-ethyl-acetate or NH3-methanol |
| Non-polar extraction on a polymer phase | EN | Drinking, ground and surface water | Polar contaminants: pesticides, phenols, explosives, anilines | Ethyl acetate, methanol, acetonitrile:methanol (1:1) |
| Non-polar extraction on a polymer phase | EN | Body fluids (not for in vitro) | Pharmaceuticals | Acetonitrile, methanol |
| Medium polar extraction of environmental pollutants | Florisil® | Waste/ground/drinking water, soil samples | Herbicides, pesticides, PCBs, PCPs, dioxins, phenols, nitro compounds, HCHs | n-Hexane, dichloromethane |

| Description | Qty. | Mfg. Cat. No. |
|--|------|---------------|
| LiChrolut® EN (40 - 120 µm) | | |
| 200 mg/3 mL | 30 | 1.19693.0001 |
| 200 mg/3 mL* | 30 | 1.19870.0001 |
| 500 mg/6 mL | 30 | 1.19691.0001 |
| 200 mg/6 mL | 30 | 1.19941.0001 |
| LiChrolut® EN / RP-18 (top) | | |
| 100/200 mg/6 mL | 30 | 1.19912.0001 |
| LiChrolut® Florisil® (150 - 250 µm) | | |
| 1000 mg/6 mL | 30 | 1.19127.0001 |
| LiChrolut® RP-18 (40 - 63 µm) | | |
| 100 mg/1 mL | 100 | 1.19855.0001 |
| 200 mg/3 mL | 50 | 1.02014.0001 |
| 500 mg/3 mL | 50 | 1.02023.0001 |
| 500 mg/6 mL | 30 | 1.19687.0001 |
| 1000 mg/6 mL | 30 | 1.02122.0001 |
| 2000 mg/6 mL | 30 | 1.19686.0001 |

| Description | Qty. | Mfg. Cat. No. |
|---------------------------------------|------|---------------|
| LiChrolut® RP-18e (40 - 63 µm) | | |
| 200 mg/3 mL | 50 | 1.19847.0001 |
| 500 mg/3 mL | 50 | 1.19849.0001 |
| LiChrolut® SCX (40 - 63 µm) | | |
| 200 mg/3 mL | 50 | 1.02016.0001 |
| 500 mg/3 mL | 50 | 1.02022.0001 |
| LiChrolut® TSC (40 - 63 µm) | | |
| 300 mg/3 mL | | 1.19767.0001 |

*glass SPE tube

Replace Classical LLE with EXtrelut® NT

SLE: Emulsion-Free Supported-Liquid Extractions

Classical liquid-liquid extraction (LLE) using a separation funnel is often associated with certain disadvantages: Formation of emulsion, poor phase separation, high solvent consumption, low degree of automation and high personnel costs. EXtrelut® NT simplifies liquid-liquid extraction by replacing separation funnels. Using a single step is more efficient and provides solvent, material, and time savings in comparison to classical funnel separation.

Specifications of EXtrelut® NT

| | | | |
|------------------------------------|---|-------|---------------------------------|
| Characteristics | Specially processed, wide-pore diatomaceous earth with a high pore volume | | |
| | Chemically inert Naturally occurring product | | |
| Capacity limit with aqueous sample | EXtrelut® NT1 | 1 mL | without any breakthrough |
| | EXtrelut® NT3 | 3 mL | |
| | EXtrelut® NT20 | 20 mL | |
| pH range | pH 1-10 | | |
| Uniform batch-to-batch quality | | | |

Benefits of EXtrelut® NT over LLE

- Minimal solvent usage
- Simple method
- Higher sample capacity and throughput
- Emulsion free extracts
- Higher purity, suitable for trace analysis

EXtrelut® NT SLE sorbent is extremely versatile and can be used for biological samples, water analysis, food and beverage, and environmental applications. Any LLE of aqueous samples can be easily replaced with EXtrelut® NT supported liquid extraction.

With its easy-to-use working principle a higher recovery and cleaner extraction can be achieved. The aqueous sample is simply applied to the LLE of aqueous samples. It distributes itself in the form of a thin film over the chemically inert matrix and thus acts as a stationary phase.

Subsequently, elution takes place using organic solvents that are non miscible with water, solvents like e.g. diethyl ether, ethyl acetate or halogenated hydrocarbons. All the



lipophilic substances are extracted from the aqueous into the organic phase. During this process the aqueous phase remains on the stationary phase. The eluate is free from emulsions and can be evaporated for further analysis.

| 1 mL | 3 mL | 20 mL |
|---------------|---------------|----------------|
| EXtrelut® NT1 | EXtrelut® NT3 | EXtrelut® NT20 |

Maximum aqueous sample capacity

The capacity of EXtrelut® NT pre-packed columns for aqueous samples are specified by the designation

Significantly smaller samples must be appropriately diluted. If larger volumes are applied, the columns are overloaded; water breaks through into the solvent. Elution is carried out with 2-3 times the sample volume. The liquid may simply be allowed to run through the column by gravity. The column outlet cannula regulates the solvent flow appropriately.

Important EXtrelut® NT extraction parameters

| EXtrelut NT® extraction columns | Outlet cannulae | Maximum sample volume (mL) | Waiting period (min) | Recommended elution volume (mL) |
|---------------------------------|-----------------|----------------------------|----------------------|---------------------------------|
| EXtrelut® NT1 | 0.60 x 30 mm | 1 | 5 – 10 | 6 |
| EXtrelut® NT3 | 0.60 x 30 mm | 3 | 5 – 10 | 15 |
| EXtrelut® NT20 | 0.70 x 30 mm | 20 | 10 – 15 | 40 |

1. In order to prevent water breaking through the sample, don't overload the column.
2. Shorter waiting times can affect the recoveries adversely.
3. The recommended sample volumes must be adhered to. Solutions of smaller volumes must be diluted to give indicated volumes.

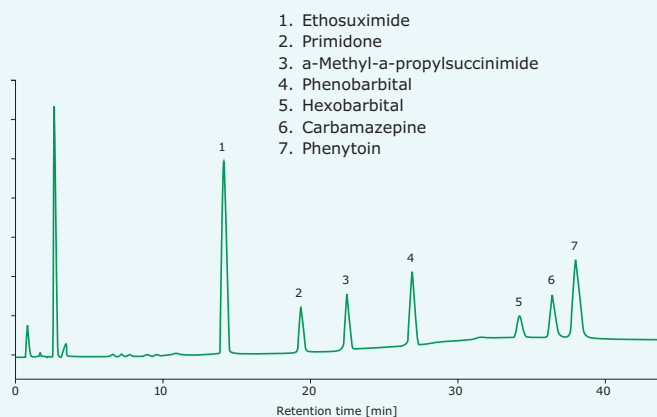
Application: HILIC separation of antiepileptic drugs (AEDs) in serum after Extrelut® NT SLE

Extrelut® NT has been used for quite some time within research, for the sample preparation of urine, whole blood, plasma, serum, gastric juice, liquor, amniotic fluid, feces, animal and plant tissue. Other applications are in the areas of environmental and residue analysis,

e.g. the analysis of industrial, domestic and waste water. The fractionated elution of acidic and basic substances (e.g. drugs and their metabolites) from body fluids is also possible.

Figure 21. HILIC Separation of Antiepileptic Drugs (AEDs) After EXTrelut® NT SLE Cleanup

| | | | |
|---------------|---|-----|-----|
| HPLC: | LaChrom® system | | |
| column: | LiChrospher® RP-select B (5 µm) LiChroCART® 250-4 | | |
| mobile phase: | A: Water LiChrosolv® Acetonitrile LiChrosolv® (1+1) | | |
| | B: Water LiChrosolv® | | |
| gradient: | Time [min] | % A | % B |
| | 0 | 10 | 90 |
| | 30 | 60 | 40 |
| | 44 | 60 | 40 |
| | 44.1 | 100 | 0 |
| | 50 | 100 | 0 |
| | 51 | 10 | 90 |
| | 75 | 10 | 90 |
| flow: | 1 mL/min | | |
| temperature: | 30 °C | | |
| detection: | UV 205 nm | | |



Determination of antiepileptic drugs (AEDs) in serum

500 µL serum
500 µL phosphate buffer*



Apply in sequence onto the column

Extrelut® NT1



Wait 8 minutes

1 mL dichloromethane / 2-propanol (9+1)



Wait 10 minutes then elute with

6 mL dichloromethane / 2-propanol (9+1)



Evaporate to dryness under nitrogen stream

Redissolve residue in 1 mL of methanol



Inject 10 µL into HPLC column

* 17.6 g NaH₂PO₄, 4.5 g Na₂HPO₄ · 2 H₂O, 1.5 g NaN₃, dissolve in 1 L water (pH 6.0-6.1)

Recoveries [mean values N = 3]

| | | |
|------------------------------|----------|-------------------|
| Ethosuximide* | 14.1 min | 84 ± 7 % |
| Primidone | 19.4 min | 100 ± 2 % |
| a-Methyl-a-propylsuccinimide | 22.5 min | Internal standard |
| Phenobarbital | 26.9 min | 96 ± 2 % |
| Hexobarbital | 34.2 min | 99 ± 2 % |
| Carbamazepine | 36.4 min | 97 ± 1 % |
| Phenytoin | 38.0 min | 100 ± 1 % |

*Ethosuximide is volatile on evaporation



Extrelut® NT collection tube for
Extrelut® NT1 and Extrelut® NT3
glass columns

EXTrelut® NT pre-packed columns

| Description | Qty. | Mfg. Cat. No. |
|---|-------------|---------------|
| EXTrelut® NT1 glass columns for 0.1 to 1 mL sample solution | 100 columns | 1.15094.0001 |
| EXTrelut® NT3 glass columns for 1 to 3 mL sample solution | 50 columns | 1.15095.0001 |
| EXTrelut® NT20 polyethylene columns including special outlet cannulae for up to 20 mL sample solution | 25 columns | 1.15096.0001 |

EXTrelut® NT packing material

| Description | Qty. | Mfg. Cat. No. |
|---|---------|---------------|
| EXTrelut® NT bulk packing for preparing large-volume columns | 1 kg | 1.15092.1000 |
| EXTrelut® NT refill packs for refilling 50 EXTrelut® NT20 columns (incl. replacement filters) | 50 bags | 1.15093.0001 |

EXTrelut® NT accessories

| Description | Qty. | Mfg. Cat. No. |
|---|------------|---------------|
| EXTrelut® NT accessories cannulae .60/30 with Luer tip for EXTrelut® NT1 and EXTrelut® NT3 | 100 pieces | 1.15373.0001 |
| EXTrelut® NT collection tubes with tapered bottom and screw cap (normal capacity 15 mL) for EXTrelut® NT1 and EXTrelut® NT3 | 30 pieces | 1.15622.0001 |
| Replacement filter for EXTrelut® NT1 (10 mm Ø) | 100 pieces | 1.14236.0001 |
| Replacement filter for EXTrelut® NT3 (15 mm Ø) | 100 pieces | 1.14237.0001 |
| Replacement filter for EXTrelut® NT20 (24 mm Ø) | 50 pieces | 1.14567.0001 |



EXTrelut® NT – Packing Material

Specialty Products for Environmental Analysis

Supelclean™ Coconut Charcoal SPE Tube for Nitrosamines in Drinking Water

- Developed specifically for EPA Method 521 – Nitrosamines in Drinking Water
- Activated coconut charcoal stationary phase – particle size: 80/120 mesh
- Quality controlled for low fines and nitrosamine recovery

| Description | Qty. | Mfg. Cat. No. |
|---|------|---------------|
| Supelclean™ Coconut Charcoal SPE Tube, 2 g/6 mL | 30 | 57144-U |
| Female Luer Coupler | 20 | 21015 |
| Male Luer Coupler | 20 | 25064-U |

Supelclean™ Sulfoxide SPE for PCB's from Transformer, Waste and Mineral Oil

- Developed for the extraction of polychlorinated biphenyls (PCBs) from transformer, waste and mineral oil
- Silica-bonded sulfoxide (-SO) phase
- PCB retention facilitated by interaction between the SPE phase's electrophilic sulfur atom and the pi-electron cloud formed from aromatic rings inherent with PCBs
- Simple and efficient sample prep method for identifying PCBs at quantitation limits of 0.5 ppm



| Description | Qty. | Mfg. Cat. No. |
|--|------|---------------|
| Supelclean™ Sulfoxide Glass SPE Tube, 6 g/20 mL | 5 | 55252-U |
| Supelclean™ Sulfoxide SPE, 3 g/6 mL | 30 | 55253-U |
| Supelclean™ Sulfoxide, Bulk, 100 g | 1 | 55254-U |
| Empty Glass SPE Tube (17 mm I.D. x 137 mm with PE frit, 20 mL, with PE frit, luer cap, and screw-top cap | 5 | 55255-U |
| Disposable PTFE liners | 100 | 57059 |
| Large volume reservoir (25 mL) for 6 mL SPE tubes, PP | 30 | 54258-U |
| Large volume reservoir (25 mL) for 6 mL SPE tubes, PTFE | 3 | 54259-U |

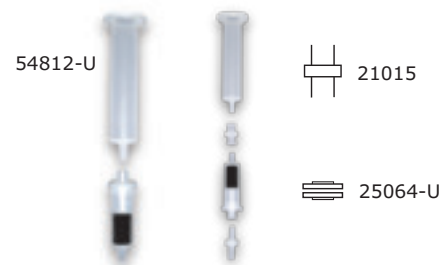
Supelclean™ ENVI-Carb™ Plus Reversible SPE for Highly Polar Compounds from Aqueous Samples

- Spherical carbon particles (carbon mol sieve) developed for the SPE of highly polar compounds from aqueous samples as drinking or ground water
- Offers extreme affinity to organic polar and non-polar compounds from both non-polar and polar matrices when used under reversed-phase conditions.
- Strong high surface spherical particles which are less friable (fines) than traditional graphitized carbon blacks
- When used in conjunction with an SPE vacuum manifold, a male luer coupler (25064-U), female luer coupler (21015) and empty SPE tube(s) are required but not included.

Examples of highly polar compounds recovered

- Acephate (LogPo/w: -0.85)
- Phenol (LogPo/w: 1.51)
- 1,4-dioxane (LogPo/w: -0.27)
- Oxamyl (LogPo/w: -1.2)


| Description | Qty. | Mfg. Cat. No. |
|---|------|---------------|
| Supelclean™ ENVI-Carb™ Plus Reversible SPE Tube, 0.4 g/1 mL | 30 | 54812-U |
| Female Luer Coupler | 20 | 21015 |
| Male Luer Coupler | 20 | 25064-U |



Specialty Products for Pesticide Analysis

Unlike typical “bind and elute” SPE practices, the modern strategy for SPE cleanup prior to routine multi-residue pesticide analysis is removal/trapping of the majority of the matrix by the sorbent phase, while the analytes of interest pass through. This results in a purified eluate. The use of packed SPE tubes, often with 2 layers of sorbent, is common. Likewise,

the “QuEChERS” approach (pg. 35) using bulk SPE materials has been incorporated into a number of methods. In all cases, the purity and the efficiency of the adsorbents used are the key to reliable and reproducible pesticide determination. With expertise in particle technology, we provide quality SPE products.

| | |
|--|---|
| Supelclean™ Ultra | <ul style="list-style-type: none"> • Designed for the cleanup of extracts of difficult matrices such as dry commodities (tea, spices, coffee, etc.) • Dual layer SPE tube contains a mixture of PSA/C18 and graphitized, spherical carbon (upper layer), and zirconia-coated silica (bottom layer) • PSA removes acidic interferences, C18 retains some hydrophobic interferences, and specialized carbon removes pigments while allowing for better recoveries of compounds with planar structures • Zirconia-coated silica (Z-Sep) removes oily residues and provides additional pigment removal |
| Supel™ Sphere Carbon/NH₂ | <ul style="list-style-type: none"> • SPE tube packed entirely with spherical, non-friable particles • Improved flow characteristics and faster flow for gravity filtration • Reduced susceptibility to the formation of fines • Dual layer SPE tube contains both spherical carbon (upper layer) and spherical silica-aminopropyl phase (lower layer), SPE sorbents are separated by a PE frit • Developed to offer superior cleanup when conducting multi-residue pesticide analysis from food • Carbon has a strong affinity toward planar molecules, and can isolate/remove pigments (eg., chlorophyll and carotinoids) and sterols commonly present in foods and natural products • Aminopropyl (NH₂) retains fatty acids, organic acids, and some polar pigments and sugars common in food matrices |
| ENVI-Carb™-II/ PSA |  <ul style="list-style-type: none"> • Dual layer SPE tube that contains both Supelclean™ ENVI-Carb™-II (upper layer) and PSA (lower layer) SPE sorbents (separated by PE frit) • Developed to offer superior cleanup when conducting multi-residue pesticide analysis in food (e.g., fruits, vegetables, etc.) • ENVI-Carb™-II a graphitized non-porous carbon (100/140 mesh, surface area 100 m²/g) that has a strong affinity towards planar molecules, and has been quality controlled specifically for the isolation/removal of pigments (e.g., chlorophyll and carotinoids) and sterols commonly present in fruits, vegetables and other natural products • Supelclean™ PSA is a polymerically bonded, ethylenediamine-N-propyl phase that contains both primary and secondary amines • Supelclean™ PSA has a strong affinity and high capacity for fatty acids, organic acids, and some polar pigments and sugars • Tested for superior cleanliness using GC/FID and GC/MS |
| ENVI-Carb™-II/ SAX/PSA | <ul style="list-style-type: none"> • Tri-layer SPE tube that contains Supelclean™ ENVI-Carb™-II (upper layer), SAX (middle layer) and PSA (lower layer) SPE sorbents (separated by PE frit) • Developed to offer superior cleanup when conducting multi-residue pesticide analysis in food (e.g., fruits, vegetables, etc.) • ENVI-Carb™-II is a graphitized non-porous carbon (100/140 mesh, surface area 100 m²/g) that has a strong affinity towards planar molecules, and has been quality controlled specifically for the isolation/removal of pigments (e.g., chlorophyll and carotinoids) and sterols commonly present in fruits, vegetables and other natural products • Supelclean™ PSA has a strong affinity and high capacity for fatty acids, organic acids, and some polar pigments and sugars • Supelclean™ SAX offers additional ion-exchange capacity for removing matrix components that may induce ion-suppression or enhancement during GC analysis |
| SAX/PSA | <ul style="list-style-type: none"> • Dual layer SPE tube that contains both Supelclean™ SAX (upper layer) and PSA (lower layer) SPE sorbents (separated by PE frit) • Supelclean™ SAX is a quarternary amine, Cl⁻ counter-ion • Supelclean™ PSA is a polymerically bonded, ethylenediamine-N-propyl phase that contains both primary and secondary amines • Ideal for removing matrix components (fatty acids, organic acids, polar pigments and some sugars) when conducting multi-residue pesticide analysis in foods • In compliance with Luke and Luke II methods that use SPE to reduce matrix induced ion-suppression and enhancement when conducting GC analysis of pesticides in food |
| ENVI-Carb™ | <ul style="list-style-type: none"> • Surface Area: 120 m²/g, Particle Size:100/400 mesh • Extreme affinity for organic polar and non-polar compounds from both non-polar 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 dispersion into solid phase pores • Independent investigators have found ENVI-Carb™ extremely useful for the rapid sample preparation of over 200 pesticides from various matrices including ground water, fruits and vegetables |
| PSA |  <ul style="list-style-type: none"> • 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 (NH₂) in terms of selectivity, but has a much higher capacity due to presence of secondary amine (0.98-1.05 meq/g) • Strong affinity and high capacity for removing fatty acids, organic acids, and some polar pigments and sugars when conducting multi-residue pesticide analysis in foods • Has been shown to significantly reduce matrix-enhancement effects encountered during the GC analysis of food products • Bidendate nature of ligands allow for chelation |

Supelclean™ Ultra

Supelclean™ Ultra solid phase extraction (SPE) cartridges were designed for the cleanup of extracts of difficult matrices such as dry commodities (tea, spices, coffee, etc.) prior to pesticide residue analysis, typically performed by GC/MS/MS and LC/MS/MS. These types of samples can contain highly concentrated pigments and oils, which may not be sufficiently cleaned using a standard QuEChERS cleanup. With little solvent usage, Ultra cartridges provide a cleaner extract and improved recovery of planar pesticides over traditional SPE cartridges without the use of toluene. By removing problematic interferences, these cartridges enable analysts to achieve detection of analytes at the ppb level.

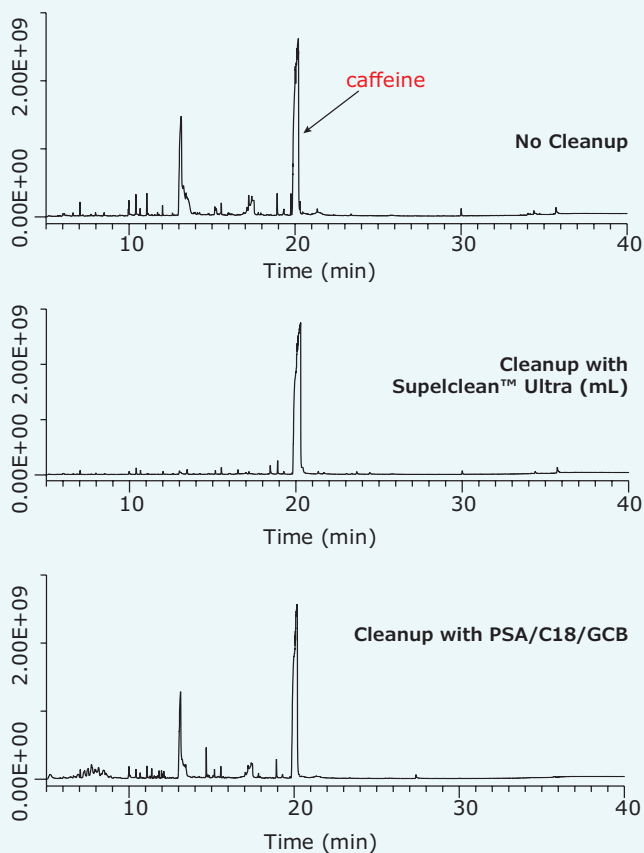
In a recent study, green tea was spiked at 5 and 50 ng/g and extracted using QuEChERS. Cleanup using a 1 mL Supelclean™ Ultra 2400 cartridge was then compared with QuEChERS cleanup using PSA/C18/GCB. The final extracts were analyzed by LC/MS/MS and GC/MS/MS. Performance of the cleanups was compared with regards to background and pesticide recoveries.

Figure 22 shows that Supelclean™ Ultra 2400 SPE was found to provide lower background than QuEChERS cleanup using PSA/C18/GCB. This allowed for the analysis of more pesticides at lower levels. These cartridges are advantageous because they use little solvent, and do not require the use of toluene in the elution solvent to release planar pesticides.

Supelclean™ Ultra SPE Products

| Description | Qty. | Mfg. Cat. No. |
|--|------|----------------|
| Supelclean™ Ultra 2400 (2 beds) | | |
| 120 mg PSA, C18, spherical carbon mix/100 mg Z-Sep, 1 mL | 108 | 52779-U |
| 270 mg PSA, C18, spherical carbon mix/225 mg Z-Sep, 3 mL | 54 | 54281-U |

Figure 22. GC/MS Scan Analyses of Green Tea Extracts



Supel™ Sphere Carbon/NH₂

Features and Benefits

- SPE tube packed entirely with spherical, non-friable particles
- Improved flow characteristics and faster flow for gravity filtration use
- Reduced susceptibility to the formation of fines
- Carbon removes pigments and sterols, commonly present in many food and natural products
- Aminopropyl (NH₂) removes organic acids, polar pigments and sugars

Spherical SPE Materials Optimize Flow and Increase Throughput

The demand for SPE cartridges with improved flow characteristics and reduced susceptibility to the formation of fines has led to the development of a family of SPE tubes packed entirely with spherical, non-friable particles. The Supel™ Sphere Carbon/NH₂ dual layer SPE tube contains both spherical carbon particles and spherical aminopropyl (NH₂) modified silica. It was developed to offer superior flow characteristics when conducting cleanup for multi-residue pesticide analysis from food.

Supel™ Sphere Carbon/NH₂ for Analysis of Pesticide Residues in Spinach

In a study comparing Supel™ Sphere Carbon/NH₂ with current products containing irregular materials, results illustrated that Supel™ Sphere Carbon/NH₂ removed as much color and background, and exhibited faster and more consistent flow than cartridges containing irregular materials, providing pesticide recovery similar to that of other dual layer SPE cartridges. Improved flow characteristics and GC/MS background is illustrated in Figures 23 and 25.

Figure 23. Flow Comparison Test

Timed Gravity Elution of Solvent (25 mL) from Dual-Layer Cartridges. Average Flow n = 5.

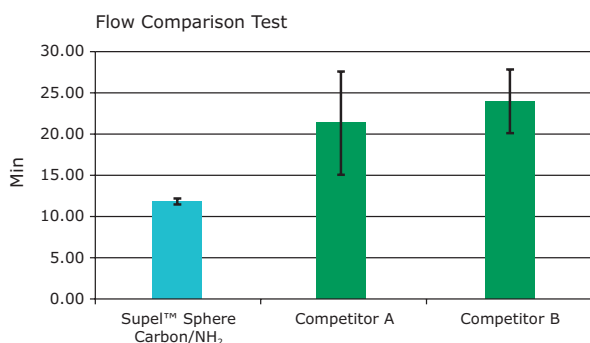


Figure 24. Supel Sphere Cartridge

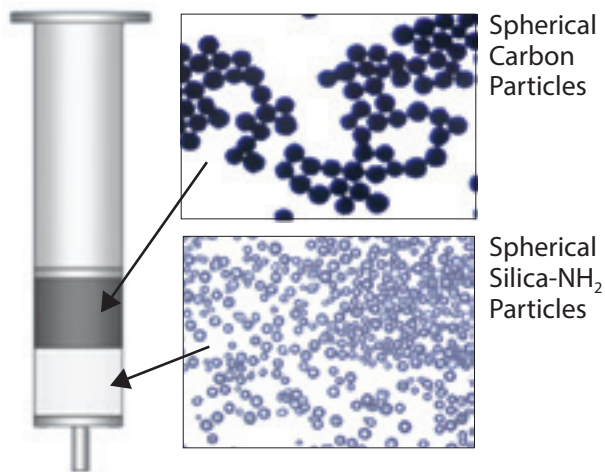
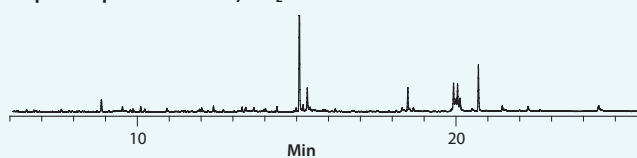


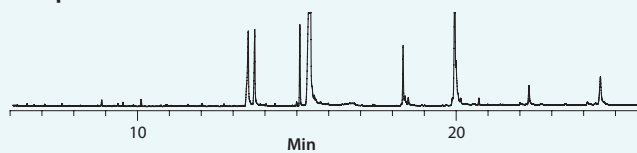
Figure 25. GC/MS Comparison of Cleaned Spinach Extracts

column: SLB®-5 ms, 20 m x 0.18 mm I.D., 0.36 µm (28576-U)
oven: 70 °C (2 min), 15 °C/min to 325 °C (5 min)
inj. temp.: Programmed, 60 °C (0.28 min), 600 °C/min to 325 °C (5 min)
carrier gas: helium, 1 mL/min constant
detector: MS, SIM mode
injection: 10 µL LVI, PTV solvent vent, rapid injection speed; split vent flow: 100 mL/min (5 psi) until 0.28 min, 60 mL/min at 2.78 min
liner: 4 mm I.D., split/splitless type, single taper FocusLiner™ design (wool packed)

Supel™ Sphere Carbon/NH₂



Competitor



| Description | Qty. | Mfg. Cat. No. |
|---|------|---------------|
| Supel™ Sphere Carbon/NH ₂ 500 mg/500 mg, 6 mL | 30 | 54283-U |

Supel™ QuE (Dispersive SPE) for “QuEChERS” Method

Quick and Simple Cleanup for Pesticide Residue Analysis

The “QuEChERS” method (Quick, Easy, Cheap, Effective, Rugged, and Safe), has emerged as a sample prep technique popular in the area of multi-residue pesticide analysis in food and agricultural products, and is formalized in the EN15662:2008 and AOAC 2007.01 Method.

In QuEChERS methodology, food/agricultural samples are first extracted with an aqueous miscible solvent (e.g., acetonitrile) in the presence of high amounts of salts (e.g., sodium chloride and magnesium sulfate) and/or buffering agents (e.g., citrate) to induce liquid phase separation and stabilize acid and base labile pesticides, respectively. Upon shaking and centrifugation, an aliquot of the organic phase is subjected to further cleanup using SPE. Unlike traditional methods using SPE tubes, in QuEChERS methodology, cleanup is facilitated by mixing bulk amounts of SPE (e.g., Supelclean™ PSA, ENVI-Carb™, and/or Discovery® DSC-18) with the extract. After sample cleanup, the mixture is centrifuged and the resulting supernatant can either be analyzed directly or can be subjected to further minor treatment before analysis.

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

For more information, visit SigmaAldrich.com/quechers



Features and Benefits

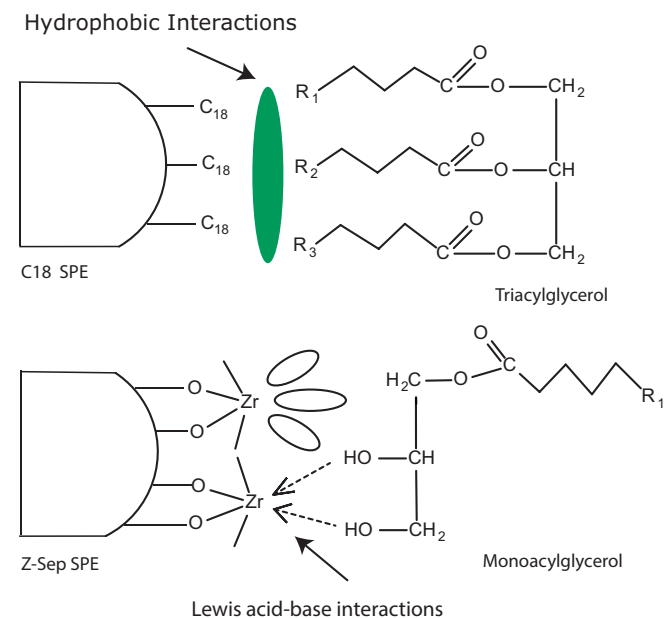
- Efficient and economic sample cleanup
- Pre-weighed amounts of sorbents and salts save labor and time
- High purity reagents
- Convenient and reliable in ready-to-use 15 mL, 12 mL and 2 mL centrifuge tubes

Supel™ QuE Z-Sep: Fat Removal in Difficult Matrices

The patent-pending zirconia-coated silica particles of Supel™ QuE Z-Sep sorbents selectively remove more fat and color from sample extracts than traditional phases for QuEChERS methods. Lipid retention is based on two synergetic interactions: the interaction between the polar group of the lipid and the proprietary bonded zirconia (Z-Sep) group of the sorbent as well as the interaction between the hydrophobic chains of the lipid and the hydrophobic group of the sorbent (either that of the C18 or Z-Sep+). Supel™ QuE Z-Sep/C18, a combination of Discovery® DSC-18 and Z-Sep particles, is recommended for samples containing <15% fat. Supel™ QuE Z-Sep+, a C18 & zirconia dual bonded silica, is recommended for cleanup of samples containing >15% fat. Supel™ QuE Z-Sep is recommended for the analysis of hydrophobic analytes in fatty matrices.

- Significantly diminishes fatty matrix interferences and various colors
- 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

Figure 26. Interactions of Supel™ QuE Z-Sep and C18



Analysis of Pesticides in Avocado using Z-Sep+ SPE Sorbent in QuEChERS Method for Sample Cleanup

In a recent experiment examining the cleanup of avocado extracts prior to pesticide residue analysis, the Z-Sep+ sorbent showed improved cleanup over PSA/C18, as illustrated in the bar chart below. The Z-Sep+ cleanup shows the lowest mass of remaining extractables after cleanup of 1.44 g of avocado. In addition, as shown in the graph below, Z-Sep+ showed improved analyte recovery over PSA/C18.

Figure 27. Total Extractables

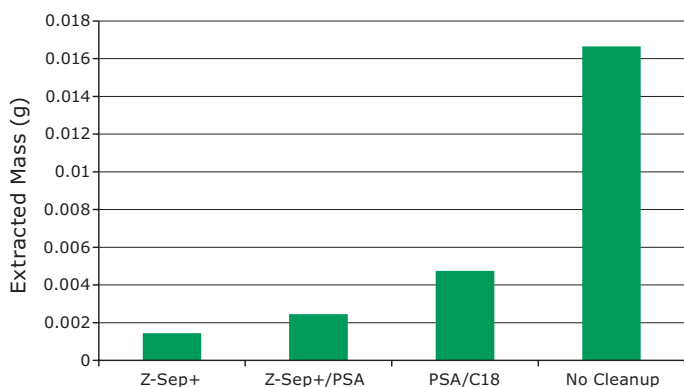
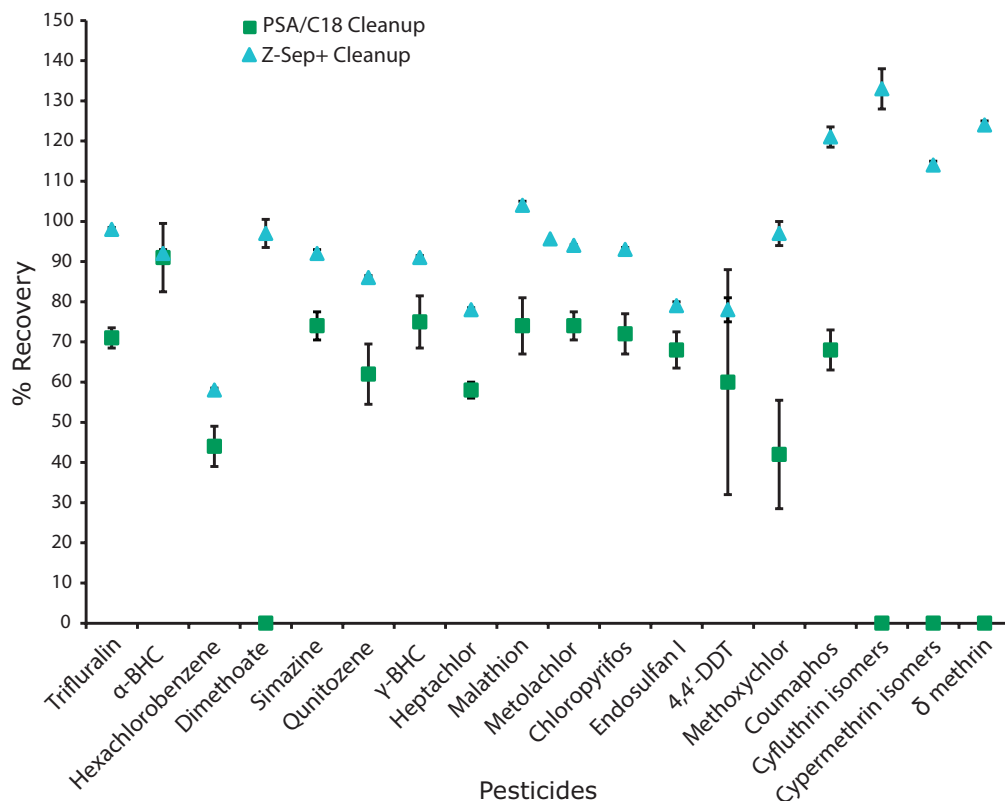


Figure 28. Analyte Recovery of Selected Pesticides from Avocado



- Z-Sep+ showed higher recovery overall.
- PSA/C18: matrix interference prevented analysis of cyfluthrin, cypermethrin and deltamethrin.
- Z-Sep+ showed better reproducibility than PSA/C18

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. As mentioned, 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.

Learn more at [SigmaAldrich.com/verde](https://www.sigmaaldrich.com/verde)

Supel™ QuE Products for QuEChERS and Related Products

Pre-Packed dSPE Tubes

| Description | Qty. | Mfg. Cat. No. |
|--|------|---------------|
| EN15662:2008 (15 mL centrifuge tubes, shaker compatible) | | |
| Supel™ QuE PSA (EN) Tube, 15 mL 150 mg Supelclean™n PSA, 900 mg MgSO ₄ | 50 | 55437-U |
| Supel™ QuE PSA/C18 (EN) Tube, 15 mL 150 mg Supelclean™ PSA, 150 mg Discovery® DSC-18, 900 mg MgSO ₄ | 50 | 55439-U |
| Supel™ QuE PSA/ENVI-Carb™ (EN) Tube 1, 15 mL 150 mg Supelclean™ PSA, 15 mg Supelclean™ ENVI-Carb™, 900 mg MgSO ₄ | 50 | 55446-U |
| Supel™ QuE PSA/ENVI-Carb™ (EN) Tube 2, 15 mL 150 mg Supelclean™ PSA, 45 mg Supelclean™ ENVI-Carb™, 900 mg MgSO ₄ | 50 | 55464-U |
| EN15662:2008 (12 mL centrifuge tubes) | | |
| Supel™ QuE Citrate (EN) Tube, 12 mL 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, 12 mL 4 g MgSO ₄ , 5 g NaBicarbonate, 1 g NaCl, 0.5 g NaCitrate dibasic sesquihydrate, 1 g NaCitrate tribasic dihydrate | 50 | 55237-U |
| Supel™ QuE PSA (EN) Tube, 12 mL 150 mg Supelclean™ PSA, 900 mg MgSO ₄ | 50 | 55228-U |
| Supel™ QuE PSA/C18 (EN) Tube, 12 mL 150 mg Supelclean™ PSA, 150 mg Discovery® DSC-18, 900 mg MgSO ₄ | 50 | 55229-U |
| Supel™ QuE PSA/ENVI-Carb™ (EN) Tube 1, 12 mL 150 mg Supelclean™ PSA, 15 mg Supelclean™ ENVI-Carb™, 900 mg MgSO ₄ | 50 | 55230-U |
| Supel™ QuE PSA/ENVI-Carb™ (EN) Tube 2, 12 mL 150 mg Supelclean™ PSA, 45 mg Supelclean™ ENVI-Carb™, 900 mg MgSO ₄ | 50 | 55233-U |
| EN15662:2008 (2 mL centrifuge tubes) | | |
| Supel™ QuE PSA (EN) Tube, 2 mL 25 mg Supelclean™ PSA, 150 mg MgSO ₄ | 100 | 55172-U |
| Supel™ QuE PSA/C18 (EN) Tube, 2 mL 25 mg Supelclean™ PSA, 25 mg Discovery® DSC-18, 150 mg MgSO ₄ | 100 | 55173-U |
| Supel™ QuE PSA/ENVI-Carb™ (EN) Tube 1, 2 mL 25 mg Supelclean™ PSA, 2.5 mg Supelclean™ ENVI-Carb™, 150 mg MgSO ₄ | 100 | 55174-U |
| Supel™ QuE PSA/ENVI-Carb™ (EN) Tube 2, 2 mL 25 mg Supelclean™ PSA, 7.5 mg Supelclean™ ENVI-Carb™, 150 mg MgSO ₄ | 100 | 55176-U |

| Description | Qty. | Mfg. Cat. No. |
|---|------|---------------|
| AOAC 2007.01 (15 mL centrifuge tubes, shaker compatible) | | |
| Supel™ QuE PSA (AC) Tube, 15 mL 400 mg Supelclean™ PSA, 1200 mg MgSO ₄ | 50 | 55466-U |
| Supel™ QuE PSA/C18 (AC) Tube, 15 mL 400 mg Supelclean™ PSA, 400 mg Discovery® DSC-18, 1200 mg MgSO ₄ | 50 | 55470-U |
| Supel™ QuE PSA/C18/ENVI-Carb™ (AC) Tube 1, 15 mL 400 mg Supelclean™ PSA, 400 mg Discovery® DSC-18, 400 mg Supelclean™ ENVI-Carb™, 1200 mg MgSO ₄ | 50 | 55474-U |
| AOAC 2007.01 (12 mL centrifuge tubes) | | |
| Supel™ QuE Acetate (AC) Tube, 12 mL 6 g MgSO ₄ , 1.5 g NaAcetate | 50 | 55234-U |
| Supel™ QuE PSA (AC) Tube, 12 mL 400 mg Supelclean™ PSA, 1200 mg MgSO ₄ | 50 | 55282-U |
| Supel™ QuE PSA/C18 (AC) Tube, 12 mL 400 mg Supelclean™ PSA, 1200 mg MgSO ₄ , 400 mg Discovery® DSC-18 | 50 | 55283-U |
| Supel™ QuE PSA/C18/ENVI-Carb™ (AC) Tube, 12 mL 400 mg Supelclean™ PSA, 1200 mg MgSO ₄ , 400 mg Discovery® DSC-18, 400 mg ENVI-Carb™ | 50 | 55286-U |
| AOAC 2007.01 (2 mL centrifuge tubes) | | |
| Supel™ QuE PSA (AC) Tube, 2 mL 50 mg Supelclean™ PSA, 150 mg MgSO ₄ | 100 | 55287-U |
| Supel™ QuE PSA/C18 (AC) Tube, 2 mL 50 mg Supelclean™ PSA, 150 mg MgSO ₄ , 50 mg Discovery® DSC-18 | 100 | 55288-U |
| Supel™ QuE PSA/C18/ENVI-Carb™ (AC) Tube, 2 mL 50 mg Supelclean™ PSA, 150 mg MgSO ₄ , 50 mg Discovery® DSC-18, 50 mg ENVI-Carb™ | 100 | 55289-U |
| Supel™ QuE PSA/ENVI-Carb™ (AC) Tube 50 mg Supelclean™ PSA, 150 mg MgSO ₄ , 50 mg ENVI-Carb™ | 100 | Custom |
| Specialty Products for Challenging (Fatty/Lipid containing) Matrices (2 mL centrifuge tubes) | | |
| Supel™ QuE Z-Sep Tube, 2 mL 75 mg Z-Sep | 100 | 55411-U |
| Supel™ QuE Z-Sep/MgSO ₄ Tube, 2 mL 50 mg Z-Sep, 150 mg MgSO ₄ | 100 | 55417-U |
| Supel™ QuE Z-Sep/C18 Tube, 2 mL 20 mg Z-Sep, 50 mg Discovery® DSC-18 | 100 | 55284-U |
| Supel™ QuE Z-Sep+ Tube, 2 mL 75 mg Z-Sep+ | 100 | 55408-U |
| Supel™ QuE Z-Sep+/MgSO ₄ Tube, 2 mL 50 mg Z-Sep+, 150 mg MgSO ₄ | 100 | 55414-U |

Supel™ QuE Products for QuEChERS and Related Products

| Description | Qty. | Mfg. Cat. No. |
|---|-------|---------------|
| Supel™ QuE Verde Tube, 2 mL 60 mg Z-Sep+, 50 mg Supelclean™ PSA, 10 mg Supelclean™ ENVI-Carb™ Y, 150 mg MgSO ₄ | 100 | 55447-U |
| Specialty Products for Challenging (Fatty/Lipid containing) Matrices (15 mL centrifuge tubes, shaker compatible) | | |
| Supel™ QuE Z-Sep Tube, 15 mL 500 mg Z-Sep | 50 | 55491-U |
| Supel™ QuE Z-Sep/MgSO ₄ Tube, 15 mL 300 mg Z-Sep, 900 mg MgSO ₄ | 50 | 55503-U |
| Supel™ QuE Z-Sep/C18 Tube, 15 mL 120 mg Z-Sep, 300 mg Discovery® DSC-18 | 50 | 55506-U |
| Supel™ QuE Z-Sep+ Tube, 15 mL 500 mg Z-Sep+ | 50 | 55486-U |
| Supel™ QuE Z-Sep+/MgSO ₄ Tube, 15 mL 300 mg Z-Sep+, 900 mg MgSO ₄ | 50 | 55511-U |
| Supel™ QuE Verde Tube, 15 mL 480 mg Z-Sep+, 400 mg Supelclean™ PSA, 80 mg Supelclean™ ENVI-Carb™ Y, 1200 mg MgSO ₄ | 50 | 55442-U |
| Specialty Products for Challenging (Fatty/Lipid containing) Matrices (12 mL centrifuge tubes) | | |
| Supel™ QuE Z-Sep Tube, 12 mL 500 mg Z-Sep | 50 | 55403-U |
| Supel™ QuE Z-Sep/MgSO ₄ Tube, 12 mL 300 mg Z-Sep, 900 mg MgSO ₄ | 50 | 55407-U |
| Supel™ QuE Z-Sep/C18 Tube, 12 mL 120 mg Z-Sep, 300 mg Discovery® DSC-18 | 50 | 55401-U |
| Supel™ QuE Z-Sep+ Tube, 12 mL 500 mg Z-Sep+ | 50 | 55296-U |
| Supel™ QuE Z-Sep+/MgSO ₄ Tube, 12 mL 300 mg Z-Sep+, 900 mg MgSO ₄ | 50 | 55406-U |
| Non-buffered extraction tubes (12 mL centrifuge tubes) | | |
| Supel™ QuE Non-Buffered Tube 1, 12 mL 4 g MgSO ₄ , 1 g NaCl | 50 | 55294-U |
| Supel™ QuE Non-Buffered Tube 2, 12 mL 6 g MgSO ₄ , 1.5 g NaCl | 50 | 55295-U |
| Specialty Extraction Salts | | |
| Supel™ QuE Ammonium Sulfate Tube, 12 mL 4 g Ammonium Sulfate | 1,000 | 54276-U |
| Empty Extraction Tubes (50 mL) | | |
| 50 mL empty Extraction Centrifuge Tubes | 50 | 55248-U |

Bulk Adsorbents and Salts

| Description | Qty. | Mfg. Cat. No. |
|--|-------|---------------|
| Supelclean™ PSA, bulk sorbent | 100 g | 52738-U |
| Supelclean™ ENVI-Carb™, bulk sorbent | 50 g | 57210-U |
| Discovery® DSC18, bulk sorbent | 100 g | 52600-U |
| Z-Sep+ | 20 g | 55299-U |
| Z-Sep | 20 g | 55418-U |
| MgSO ₄ (as cited in EN15662:2008) | var. | 208094 |
| Sodium citrate dibasic sesquihydrate | var. | 71635 |
| Sodium citrate tribasic dihydrate | var. | 54641 |
| Sodium chloride | var. | 71379 |
| Sodium acetate | var. | 241245 |

QuEChERS Shakers and Accessories

| Description | Qty. | Mfg. 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 | — | Z765503 |
| Multi-tube Vortexer, EU compatible Schuko plug, AC input 230 V | — | Z765511 |
| Benchmark Benchmixer™ XL Laboratory Shaker Racks | | |
| 50 mL QuEChERS Extraction Tube Shaker Rack | 1 | 55279-U |
| 15 mL QuEChERS Cleanup Tube Shaker Rack | 1 | Z765589 |
| 2 mL QuEChERS Cleanup Tube Shaker Rack | — | Z765554 |



Specialty Products for Mycotoxin Analysis

Supel™ Tox SPE Cartridges

Features and Benefits

- Removes interferences associated with mycotoxin analysis
- Basic and quick methodology requiring no additional method development
- Time associated with sample preparation is up to ten times less than that associated with immunoaffinity columns, the current industry standard
- No refrigeration required for shipping and storage of cartridges



Supel™ Tox SPE Products

| Description | Use |
|-----------------------|--|
| Supel™ Tox AflaZea | Cleanup of grains, feed, TMR samples, peanuts, peanut products, and aqueous solutions for detection of aflatoxin and zearalenone |
| Supel™ Tox DON | Cleanup of wheat, flour and corn for detection of deoxynivalenol (DON) |
| Supel™ Tox Tricho | Cleanup of grains and complex matrices for detection of Type A and B Trichothecenes |
| Supel™ Tox TrichoBind | Cleanup of grains and complex matrices for the detection and purification of Type A and B Trichothecenes |
| Supel™ Tox FumoniBind | Cleanup of whole grains and cereals for detection of fumonisin (B ₁ and B ₂) |
| Supel™ Tox OchraBind | Cleanup of whole grain and feed samples for the detection of ochratoxin A |

Supel™ Tox AflaZea, DON, Tricho apply interference removal strategy.
Supel™ Tox TrichoBind, FumoniBind and OchraBind apply Bind& Elute strategy.

Fast and Simple Cleanup for Mycotoxin Analysis

The need for a quick, simplistic sample cleanup approach prior to mycotoxin analysis has brought about a line of SPE cartridges that significantly decrease sample prep time, increase reproducibility, and are more user friendly as compared to the industry standard immunoaffinity columns (IAC). In addition, the Supel™ Tox SPE approach requires less equipment and fewer consumables, providing additional cost savings.

Table 1. Sample Cleanup Procedures Using Supel™ Tox AflaZea SPE Cartridges and Immunoaffinity Columns (n=3)

| | Immunoaffinity | Supel™ Tox AflaZea SPE Cartridge |
|--|---|---|
| Sample Prep Time (post-extraction to pre-analysis) | <ul style="list-style-type: none"> • 60 minutes • 8 samples/day (if processing 1 at a time) | <ul style="list-style-type: none"> • 6 minutes • 80 samples/day (if processing 1 at a time) |
| Ease of Use | <ul style="list-style-type: none"> • Large volumes of liquid • Controlled drop rates • Numerous complicated steps • Additional buffer salts required • Must be refrigerated, brought to room temp before use | <ul style="list-style-type: none"> • Small volumes of liquid • Vacuum filtration used • Steps few and not complicated • No additional reagents required • Column does not require special storage conditions |
| Procedure (post-extraction to analysis) | <p>Stage 1 (15 minutes)</p> <ol style="list-style-type: none"> 1. Configure manifold for waste collection 2. Add 1 mL sample to 17 mL of phosphate buffered saline, vortex 3. Uncap/mount/drain cartridges by gravity 4. Apply reservoirs, load sample onto cartridges <p>Stage 2 (15 minutes)</p> <ol style="list-style-type: none"> 1. Rinse interferences 2. Reconfigure manifold for sample collection 3. Elute/collect sample <p>Stage 3 (30 minutes)</p> <ol style="list-style-type: none"> 1. Evaporate sample to dryness 2. Reconstitute sample and vortex 3. Transfer 0.2 mL sample to vial 4. Dilute sample and vortex <p>Analysis</p> | <p>Purify and Transfer (6 minutes)</p> <ol style="list-style-type: none"> 1. Configure manifold for sample collection 2. Mount cartridges 3. Load 2 mL sample 4. Elute and collect under vacuum 5. Transfer 0.2 mL sample to vial 6. Dilute sample and vortex <p>Analysis</p> |

Application: HPLC Analysis of Aflatoxins in Raw Peanut Paste

A comparison of sample processing time, product performance, and process simplicity associated with the use of IAC and SPE cleanup methods for the analysis of aflatoxins in peanut paste is described herein. Sample purification procedures comparing cleanup with a leading brand of IAC columns to SPE cleanup using Supel™ Tox AflaZea cartridges (n=3) are summarized in **Table 1** (on previous pg). The time required for each procedure was recorded and averaged. Chromatographic analysis was performed by HPLC with fluorescence detection using a Discovery® C18 column and a KOBRA® electro-chemical cell for aflatoxin derivatization.

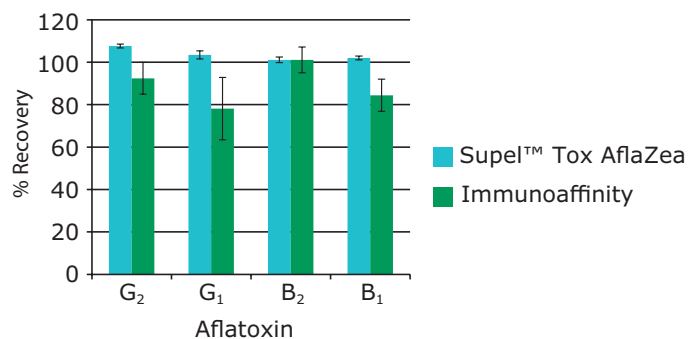
Sample Preparation (Time and Ease of Use)

As illustrated in **Table 1**, the use of the Supel™ Tox AflaZea SPE cartridges for sample cleanup was 10 times faster than that of the IAC columns. Use of the SPE cartridges eliminated the need for buffer solution, waste collection glassware, manifold reconfiguration, and equipment necessary to evaporate samples to dryness, making the SPE cartridges more user friendly than the IAC columns.

Analyte Recovery

The average % recoveries and %RSD values were compared for IAC and SPE purification techniques. **Figure 29** illustrates that Supel™ Tox AflaZea SPE cartridges gave higher analyte recoveries of B₁, G₁, B₂, and G₂ than the IAC columns used in this study. Also, as shown by the error bars, the %RSD was much lower for the SPE purification than the IAC purification, indicating that the SPE cartridges demonstrated better reproducibility than IAC for the analysis of aflatoxins in peanut paste.

Figure 29. Cleanup of Aflatoxins in Peanut Paste: Supel™ Tox AflaZea SPE Cartridges vs. Immunoaffinity Columns



Conclusion

This experiment illustrated that sample preparation using Supel™ Tox AflaZea SPE cartridges for cleanup was fast and simple compared to the IAC cleanup method. Because there were fewer steps needed to accomplish the SPE method, less variability was introduced into sample preparation, giving a more reproducible method. Also, the time associated with sample prep using SPE was far less than that associated with IAC, allowing for an ultimate increase in sample throughput. In addition, labware, reagents, and necessary equipment to perform sample preparation were minimal when using SPE. In this study, Supel™ Tox AflaZea SPE cartridges demonstrated superiority over IAC columns in terms of process simplicity, time required for sample preparation, and control of variation while maintaining the same sample cleanup performance associated with IAC purification.

| Description | Qty. | Mfg. Cat. No. |
|--|------|---------------|
| Supel™ Tox AflaZea SPE Cartridge, 6 mL | 30 | 55314-U |
| Supel™ Tox DON SPE Cartridge, 6 mL | 30 | 55316-U |
| Supel™ Tox Tricho SPE Cartridge, 6 mL | 30 | 55308-U |
| Supel™ Tox TrichoBind SPE Cartridge, LRC | 25 | 55307-U |
| Supel™ Tox FumoniBind SPE Cartridge, LRC | 25 | 55315-U |
| Supel™ Tox OchraBind SPE Cartridge, LRC | 25 | 55318-U |

Specialty Products for Analytes in Edible Oils

Supelclean™ EZ-POP NP SPE Cartridges

Features and Benefits

- Provides simultaneous extraction of a full range of polycyclic aromatic hydrocarbons (PAHs), while removing both fatty matrix and polar interferences from oil matrices
- Produces cleaner extracts and gives better overall PAH recoveries than other SPE methods
- Easier and more versatile methodology than other SPE methods, requiring fewer steps and little to no method development
- Final extracts are GC and HPLC compatible
- Yields clean extracts which can be analyzed using any MS detector

Simple, Effective Extraction of Lipophilic Persistent Organic Pollutants (POPs) from Oily Samples

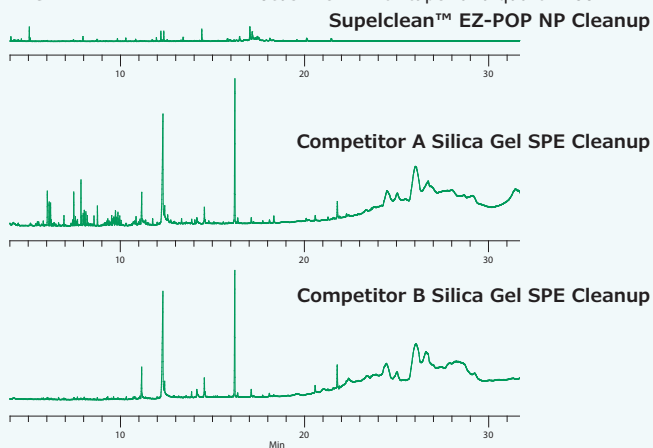
This dual-layer SPE cartridge offers superior cleanup for the extraction of non-polar POPs, specifically heavy and light PAHs, from edible oil matrices. The top Florisil® layer retains polar functional groups such as acids and alcohols. The bottom Z-Sep/C18 layer binds fatty matrix through hydrophobic interaction as well as Lewis acid-base interactions. Fatty matrix is preferentially retained by the cartridge while non-polar POPs, are washed through using acetonitrile. The resulting extract is suitable for either GC/MS or HPLC analysis.

Application: The Analysis of PAHs in Olive Oil

The Supelclean™ EZ-POP NP was compared to two competitor silica gel SPE cartridges in terms of matrix removal and analyte recovery for the extraction of

Figure 30. GC/MS Full Scan Chromatograms of Olive Oil Extract (same y axis)

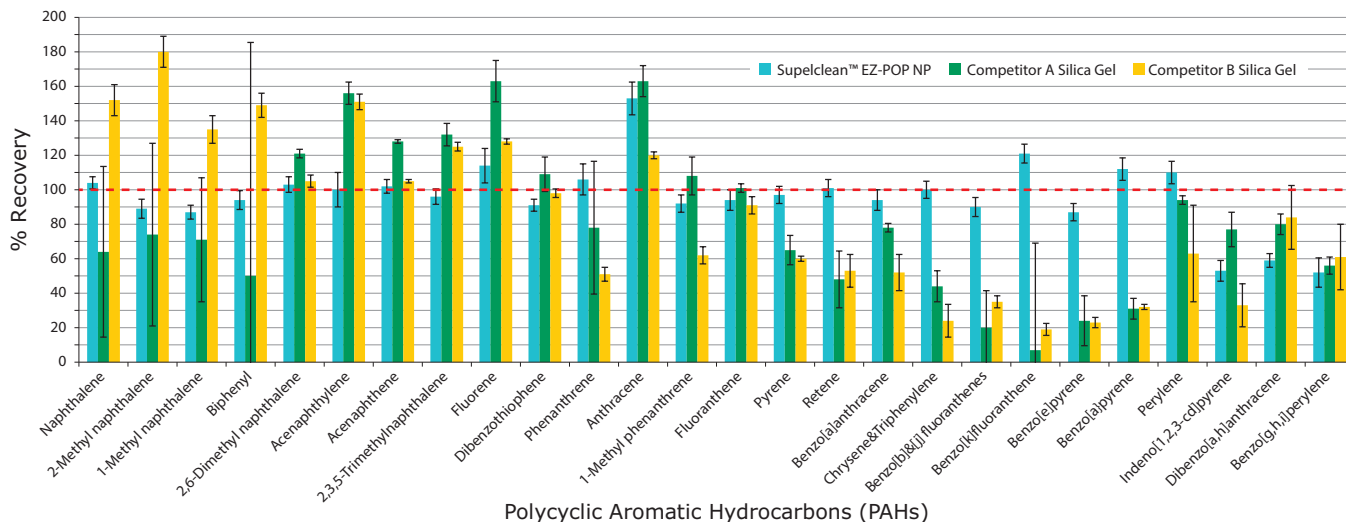
column: SLB®-5ms, 20 m x 0.18 mm I.D., 0.18 µm (28564-U)
 oven: 60 °C (1 min.), 15 °C/min. to 250 °C, 8 °C/min. to 330 °C (7 min.)
 inj. temp.: 300 °C
 carrier gas: helium, 1 mL/min constant flow
 detector: MS
 MSD interface: 330 °C
 injection: 1 µL, pulsed splitless (50 psi until 0.75 min, splitter open at 0.75 min.)
 liner: 4 mm ID FocusLiner™ with taper and quartz wool



select PAHs from olive oil. The EZ-POP NP removed more unwanted background than silica gel SPE, greatly decreasing the matrix effects (Figure 30). It produced better, more accurate, analyte recoveries than the silica gel SPE with good reproducibility (Figure 31). Thus, the Supelclean™ EZ-POP NP provides suitable matrix removal for rugged GC/MS analysis of PAHs in olive oil.

| Description | Qty. | Mfg. Cat. No. |
|-----------------------------------|------|---------------|
| Supelclean™ EZ-POP NP, 2.5 g/1 mL | 20 | 54341-U |

Figure 31. Analyte Recovery of PAHs from Olive Oil Extract (n=3)



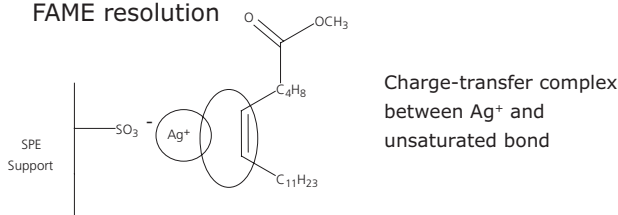
Miscellaneous Specialty Products and SPE Accessories

Discovery® Ag-Ion SPE Tubes for *cis/trans* FAME Analysis

Retention Mechanism: Normal-phase

Sample Matrix Compatibility: Organic solvents, oils, and lipids

- Developed for the fractionation of FAMEs based on degree of unsaturation and for the resolution of *cis/trans* isomers.
- Silver counter-ions are anchored onto a SCX support using a proprietary procedure to offer optimal resolution, performance and capacity.
- Each lot is tested and quality controlled for *cis/trans* FAME resolution



| Description | Qty. | Mfg. Cat. No. |
|----------------------------------|------|---------------|
| 750 mg/6 mL | 30 | 54225-U |
| 750 mg/1 mL reversible cartridge | 10 | 54226-U |

Glass SPE Tubes with PTFE Frits

A select line of our Supelclean™ SPE phase chemistries is also available in inert glass and PTFE hardware configurations.

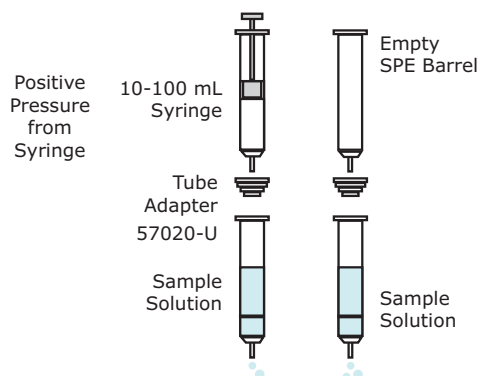


- Resistant to harsh chemicals and aggressive solvents
- Absence of leachables such as phthalates and plasticizers
- Hygroscopic adsorbents (e.g. Florisil®) can be easily heat treated/activated (e.g., 105-120 °C oven, overnight) prior to use.

| Description | Qty. | Mfg. Cat. No. |
|--|------|---------------|
| Supelclean™ ENVI-18 SPE Tube | | |
| bed wt. 500 mg, vol. 6 mL | 30 | 54331-U |
| Supelclean™ ENVI-8 SPE Tube | | |
| bed wt. 500 mg, vol. 3 mL | 27 | 57106 |
| bed wt. 500 mg, vol. 6 mL | 20 | 57107 |
| Supelclean™ LC-Florisil® SPE Tube | | |
| bed wt. 500 mg, vol. 6 mL | 30 | 54333-U |
| bed wt. 1 g, vol. 6 mL | 30 | 54334-U |
| Supelclean™ LC-Si SPE Tube | | |
| bed wt. 1 g, vol. 6 mL | 30 | 54335-U |
| Dual Layer Florisil®/Na₂SO₄ SPE Tube | | |
| bed A: 2 g (Na ₂ SO ₄), bed B: 2 g (Florisil®), vol. 6 mL | 48 | 52582-U |

Accessories

Tube Adapters



Tube adapters serve many functions:

- Stack one SPE tube on top of another to provide different selectivities
- A larger empty syringe barrel can be stacked on top of a smaller SPE tube to act as a larger load reservoir
- Adapter for positive pressure methods (e.g. from a syringe or air/N₂ line)

| Description | Qty. | Mfg. Cat. No. |
|---|------|---------------|
| SPE Tube Adapters for Polypropylene Tubes | | |
| For 1, 3, 6 mL Tubes | 12 | 57020-U |
| For 12, 20, 60 mL Tubes | 6 | 57267 |
| AutoTrace® SPE Tube Adapters* | | |
| For 3 mL Tubes | 6 | 57123 |
| For 6 mL Tubes | 6 | 57126 |
| * Allows SPE tubes to be used with AutoTrace® Automated Systems | | |
| SPE Tube Adapter for Glass Tubes | | |
| PTFE, for use with 6 mL glass SPE Tube | 24 | 504335 |

Large Volume SPE Reservoirs

Large volume SPE reservoirs are designed to increase the headspace volume of standard polypropylene SPE tubes. Because these reservoirs are designed to connect directly to the mouth of the SPE tube, they are ideal for gravity applications where increased headspace volume is required.

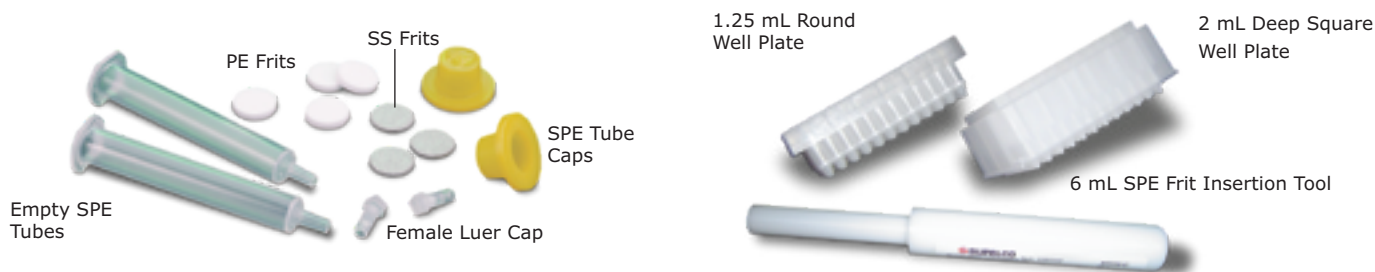


The reservoirs are designed for use with 6 mL polypropylene SPE tubes and add an additional headspace volume of 25 mL.

| Description | Qty. | Mfg. Cat. No. |
|-----------------------------------|------|---------------|
| Large Volume SPE Reservoir | | |
| Polypropylene | 30 | 54258-U |
| PTFE | 3 | 54259-U |

SPE Accessories

Empty SPE Hardware and Components



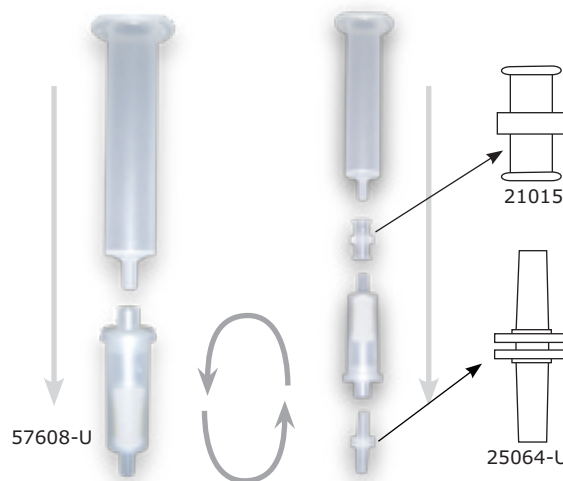
SPE Tube Components

| Description | 1 mL | 3 mL | 6 mL | 12 mL | 20 mL | 60 mL |
|---|------------------|------------------|------------------|------------------|---------------|-----------|
| Empty SPE Tubes with and without Frits | Qty. | 108 | 54 | 30 | 20 | 16 |
| Empty PP SPE Tube with PE Frits, 20 µm porosity | 57023 | 57024 | 57026 | 57176 | 57177 | 57178 |
| Empty PP SPE Tube with PE Frits, 20 µm porosity – pre-fritted with bottom frit | 54220-U (pk 100) | 54221-U (pk 100) | 54222-U (pk 100) | 54223-U (pk 100) | 57118-U | 57119-U |
| Empty PP SPE Tube (no frits) | 57240-U | 57241 | 57242 | 57179 (pk 12) | 57021 (pk 12) | 57022 |
| Empty Glass SPE Tubes with PTFE Frits, 20 µm porosity | — | — | 504394* | — | — | — |
| SPE Tube Caps (encloses top of SPE tubes) | Qty. | 108 | 54 | 30 | 20 | 20 |
| PP cap for PP SPE tubes | 52171-U | 52172-U | 52173-U | 52174-U | 52175-U | 52176-U |
| PTFE cap for glass SPE tube | — | — | 504343* | — | — | — |
| Frits for use with SPE tubes | Qty. | 216 | 108 | 60 | 40 | 32 |
| PE Frits for PP SPE tubes, 20 µm porosity | 57244 | 57180-U | 57181 | 57182-U | 57183 | 57184 |
| PTFE Frits for PP SPE tubes, 20 µm porosity | 57185 | 57186 | 57187 | 57188 | — | 57190-U |
| PTFE Frits for glass SPE tubes, 20 µm porosity | — | — | 504327 | — | — | — |
| SS Frit for PP SPE tubes, 20 µm porosity | — | — | 57246-U | — | — | — |
| SPE Frit Insertion Tool | | | | | | |
| SPE Frit Insertion Tool, pk 1 | 55217-U | 55218-U | 55219-U | 55221-U | 55224-U | 55224-U |
| SPE Frit Insertion Tool Kit (includes all 5 tools for 1, 3, 6, 12 and 20/60 mL tubes) | — | — | — | — | 55226-U | — |

PP = Polypropylene; PTFE = Polytetrafluoroethylene; SS = Stainless steel; PE = Polyethylene * Qty. of 24

Miscellaneous SPE Hardware and Accessories

| Description | Qty. | Mfg. Cat. No. |
|---|------|---------------|
| Empty Reversible SPE Tube, non-fluorous PP, w/PE frits | | |
| 0.5 mL | 50 | 57602-U |
| 1.0 mL | 50 | 57607-U |
| 2.0 mL | 50 | 57608-U |
| Empty PP Rezorian Tube Kit w/PE Frits, luer plugs and caps | | |
| 1.0 mL | 50 | 57609-U |
| 5.0 mL | 50 | 57613-U |
| Empty 96-well SPE Plates | | |
| 2 mL deep square well, w/PE frits | 1 | Inquire |
| 1.25 mL round well, w/PE frits | 1 | Inquire |
| Luer Caps, Plugs, and Couplers | | |
| Female Luer Cap, PP (caps SPE luer tips) | 12 | 57098 |
| Male Luer Plug, PP (plugs female luer fitting) | 12 | 504351 |
| Female Luer Coupler | 20 | 21015 |
| Male Luer Coupler | 20 | 25064-U |



Visiprep™ and Visiprep™ DL SPE Vacuum Manifolds

Visiprep™ SPE Vacuum Manifolds allow you to process up to 12 or up to 24 SPE tubes simultaneously. Both DL (disposable liner) and standard models are available.



12-Port Visiprep™ DL Vacuum Manifold (57044)

The Visiprep™ DL Vacuum Manifold eliminates the possibility of cross contamination when processing a new sample on the same port by employing a disposable liner that builds the complete flow path through the valve. The liner consists of a PP luer hub that attaches to the SPE tube, and a thin walled PTFE tubing that is threaded through the

SPE port. This ensures that all SPE port/valve surfaces coming in contact with the sample can be easily & conveniently replaced following each extraction.

Features and Benefits DL and Standard Models

- Screw-type valves for each SPE port for precise flow control by just turning the attached SPE tube
- Glass basin will not dissolve, fog or discolor when exposed to solvents
- Legs on stand-alone cover allows user to easily rest cover on work surface when removed from vacuum manifold
- Screw type solvent resistant vacuum bleed gauge and valve offer better sealing
- PP collection vessel rack accommodates autosampler vials, small scintillation vials, 10 and 16 mm test tubes and 1, 2, 5, and 10 mL volumetric flasks. An optional plate for 20 mL scintillation vials is available for 24-port models.

| Description | Mfg. Cat. No. |
|---|---------------|
| Visiprep™ DL Solid Phase Extraction Manifold | |
| 12-Port Model | 57044 |
| 24-Port Model | 57265 |
| Disposable valve liners, PTFE, pk. of 100 | 57059 |
| Visiprep™ Solid Phase Extraction Manifold | |
| 12-Port Model | 57030-U |
| 24-Port Model | 57250-U |



24-Port Visiprep™ Vacuum Manifold (57250-U)

Visiprep™ 5-Port Flask Manifold

The Visiprep™ 5-Port Flask Vacuum Manifold enables analysts using solid phase extraction tubes to simultaneously prepare up to 5 samples.



Unlike conventional vacuum manifolds, the Visiprep™ 5-Port Flask Manifold allows users to collect their SPE eluate directly into 50 mL round or flat bottom flasks for direct rotovap evaporation. The manifold consists of a chemical resistant 5-port cover (DL or standard available), gasket, base, a glass basin, vacuum gauge and bleed valve, 5 flow control valves, 5 replaceable solvent guide needles and a base plate that supports up to five 50 mL round or flat bottom flasks. Each port on both the standard and DL Visiprep™ models are equipped with flow control valves.

Recommended Flasks: Aldrich® single-neck flask, 50 mL, joint: ST/NS 24/40

- Round Bottom (Cat. No. Z414484)
- Flat Bottom (Cat. No. Z418773)

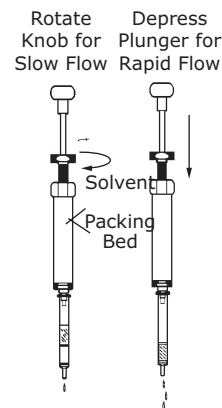
| Description | Mfg. Cat. No. |
|---|---------------|
| Visiprep™ 5-Port Flask Vacuum Manifold | |
| DL (Disposable Liner) | 57101-U |
| Standard | 57103-U |
| Visiprep™ 5-Port Vacuum Manifold Conversion Kit | |
| For converting 24-port model into DL 5- port flask model, includes DL 5-port lid and flask base plate | 57104-U |

Visi-1™ Single SPE Tube Processor

Visi-1™ processor - two rates of flow control

Our Visi-1™ Single SPE Tube Processor provides precise flow control through a single 1 mL, 3 mL or 6 mL SPE tube. There is no faster, more convenient, or more reliable method for processing one or a few samples.

Simply fill the SPE tube with the appropriate solution and attach it to the Visi-1™ processor. Remove the tube from the processor, introduce the next solution and repeat the process.



| Description | Mfg. Cat. No. |
|-----------------------------------|---------------|
| Visi-1™ Single SPE Tube Processor | 57080-U |

Preppy™ Vacuum Manifold

Simultaneously prepare up to 12 samples with our simplest and most economical manifold. The Preppy™ consists of a chemical-resistant cover and gasket, glass basin, vacuum release vent and 12 individual control valves with knurled tops and stainless steel solvent guide needles.

Two optional collection racks are available – one for 2 and 4 mL autosampler vials and the other for 15 (w/21 mm O.D.) or 40 (w/28 mm O.D.) mL vials. An optional vacuum gauge/bleed valve assembly can be installed to allow precise control of the vacuum.

| Description | Mfg. Cat. No. |
|--|---------------|
| Preppy™ Vacuum Manifold | |
| 12-Port Model | 57160-U |
| Preppy™ Replacement Parts | |
| Cover with flow control valves and solvent needle guides | 57158-U |
| Collection Vessel Racks | |
| For 2 or 4 mL vials | 57159-U |
| For 15 or 40 mL vials | 57162-U |
| Accessories | |
| Vacuum Gauge/Bleed Valve Assembly | 57161-U |

Chemical
Resistant Cover

Glass Basin



Visidry™ Drying Attachment

Designed for our Visiprep™ Vacuum Manifold, the Visidry™ Drying Attachment (57100-U) also fits our economical Preppy™ manifold.

The Visidry™ unit installs in minutes, dries up to 12 or up to 24 SPE tubes at one time and can be used with any inert gas supply. It is also useful for evaporating and concentrating recovered samples. (Gas) flow through each Visiprep port can be still independently adjusted.

57100-U



57030-U 12-Port Model
Order Separately

| Description | Qty. | Mfg. Cat. No. |
|---|------|---------------|
| Visidry™ Drying Attachment | | |
| 12-Port Model | 1 | 57100-U |
| 24-Port Model | 1 | 57124 |
| Replacement Parts for Visidry™ Drying Attachment | | |
| Control Knobs | 2 | 57095 |
| Retaining "C" Clips | 2 | 57096 |
| Female Luer Plugs | 12 | 57098 |

Replacement SPE Tube Adapters (57020-U) listed on p. 42.

NOTE: The Visidry™ drying attachment cannot be used to dry 12 mL, 20 mL, or 60 mL SPE tubes.

Visiprep™ Large Volume Samplers

Allows for easy "hands-off" transfer of large volumes of low viscosity liquid samples directly from any sample container to conventional SPE tubes (not suitable for glass tubes).

The samplers consist of 1/8" PTFE tubing with a stainless steel weight at one end and a screw-fitted SPE tube adapter on the other end. To use the sampler, the weighted end is placed in the sample container, and the tube adapter is inserted into a pre-conditioned SPE tube. Vacuum pressure delivered from the vacuum manifold is used to pull the sample through the PTFE tubing into the SPE tube where analytes of interest are concentrated on the SPE tubes prior to elution.



| Description | Qty. | Mfg. Cat. No. |
|---|------|---------------|
| Visiprep™ Large Volume Sampler | | |
| for 12 mL, 20 mL, or 60 mL SPE Tubes (3 adapters) | 1 | 57272 |
| for 3 mL or 6 mL SPE Tubes (4 adapters) | 1 | 57275 |
| Replacement Parts | | |
| 1/8" PTFE Tubes, color-coded | 4 | 57276 |
| Nuts and Ferrules, color-coded | 4 | 57277 |
| Stainless Steel Weights | 4 | 57278 |
| Tube Adapters, 1/4-28 threads | | |
| For 3 mL or 6 mL Tubes | 4 | 57273-U |
| For 12 mL, 20 mL, or 60 mL Tubes | 3 | 57274-U |

SPE Elution Rack for Gravity Feed Elution

This versatile stand-alone elution rack can be used with a variety of SPE tubes and receiving vessels, for simultaneous gravity feed extraction of up to 12 tubes. By assembling the plates in appropriate combinations, you can configure the rack to accept the following:

- 1 mL, 3 mL or 6 mL syringe barrel-type tubes
- Closed cartridge (reversible) tubes
- 5 mL or 10 mL volumetric flasks
- 2 mL or 4 mL vials
- Test tubes up to 15 mm I.D. x 10 cm



| Description | Mfg. Cat. No. |
|------------------|---------------|
| SPE Elution Rack | 21043-U |

Vacuum Manifold Replacement Parts and Accessories

| Description | Qty. | Mfg. Cat. No. |
|--|------|---------------|
| For 12-Port Manifold | | |
| Cover, 12 flow control valves, gasket ¹ | - | 57031-U |
| Cover, 12 DL flow control valves, gasket ² | - | 57029 |
| Gaskets | 2 | 57033 |
| Collection rack (base, 3 support rods, center plate, 10 mm test tube plate, 12 retaining clips) ³ | - | 57037 |
| Plate for 16 mm test tubes ³ | - | 57039 |
| Plate for 2 mL autosampler vials ³ | - | 57040-U |
| Plate for 20 mL scintillation vials | - | 57043 |
| Splash guard | - | 57045-U |
| For 24-Port Manifold | | |
| Cover, 24 flow control valves, gasket ⁴ | - | 57251 |
| Cover, 24 DL flow control valves, gasket ⁵ | - | 57266 |
| Gaskets | 2 | 57254 |
| Collection rack (base, 2 support rods, center plate, 10 mm test tube plate, 8 retaining clips) ⁶ | - | 57255 |
| Plate for 16 mm test tubes ⁶ | - | 57257 |
| Plate for 2 mL autosampler vials ⁶ | - | 57258 |
| For 12-Port or 24-Port Manifold | | |
| Valve Stem for Visiprep™ DL Vacuum Manifold | 24 | 57146-U |
| Valve Stem for Visiprep™/Preppy™ Vacuum Manifold | 24 | 57147-U |
| Flow control valves ⁷ | 2 | 57032 |
| Solvent guide needles, PTFE ^{1,8} | 12 | 57047 |
| Solvent guide needles, stainless steel ⁷ | 12 | 57036 |
| Disposable valve liners for DL versions, PTFE ^{2,5} | 100 | 57059 |
| Disposable liner flow control valves ⁹ | 2 | 57028 |
| Liner guide needles, stainless steel ^{2,10} | 12 | 57027 |
| Vacuum gauge and bleed valve | | 57035-U |
| Retaining clips for collection racks | 12 | 57041 |
| Test tubes, 10 x 75 mm ^{1,2,8,10} | 12 | 57042 |

¹ Compatible with 57030-U

² Compatible with 57044

³ Compatible with 57030-U and 57044

⁴ Compatible with 57250-U

⁵ Compatible with 57265

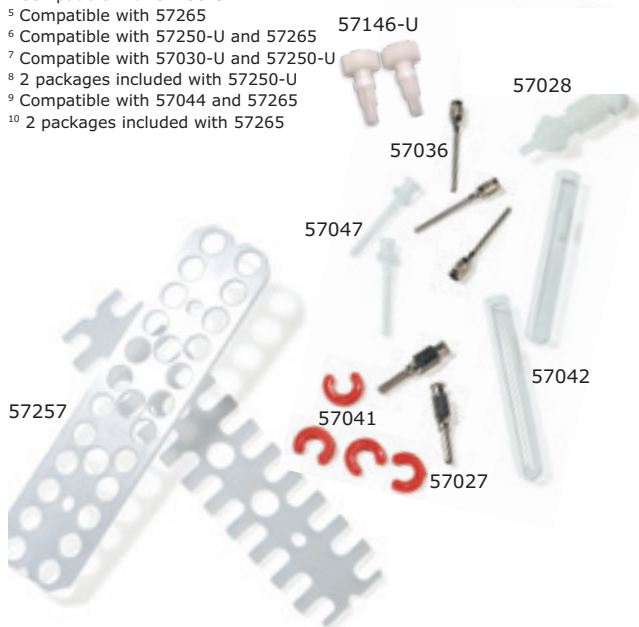
⁶ Compatible with 57250-U and 57265

⁷ Compatible with 57030-U and 57250-U

⁸ 2 packages included with 57250-U

⁹ Compatible with 57044 and 57265

¹⁰ 2 packages included with 57265



Trap Kit for SPE Vacuum Manifolds

When installed between a Visiprep™ SPE vacuum manifold and the vacuum source, a SPE Vacuum Pump Trap collects all liquids that are aspirated through the SPE tubes, preventing contamination of the vacuum pump.



The easily assembled kit contains a polypropylene filtering flask, a one-hole rubber stopper, 4" (10 cm) of polypropylene tubing and 5' (1.5 m) of red rubber vacuum hose.

| Description | Mfg. Cat. No. |
|--------------------------|---------------|
| SPE Vacuum Pump Trap Kit | 57120-U |

Vacuum Gauge / Bleed Valve Assembly

Install in-line for control of vacuum.



| Description | Mfg. Cat. No. |
|-------------------------------------|---------------|
| Vacuum Gauge / Bleed Valve Assembly | 57161-U |

Long Stem Flow Control Valves for Visiprep™ Manifolds

Equip alternate valves in your standard 12-port or 24-port Visiprep™ vacuum manifold with these long stem flow control valves if you intend to use all ports of the manifold with 12 mL, 20 mL or 60 mL tubes.

Not for use with DL manifolds.



| Description | Qty. | Mfg. Cat. No. |
|-------------------------------|------|---------------|
| Long Stem Flow Control Valves | 6 | 57048 |

Long Stem Flow Control Knobs

If you have equipped your Visiprep™ Vacuum Manifold with long stem flow control valves, these control knobs will enable you to attach the Visidry™ Drying Attachment without removing the long stem valves.

NOTE: Not to be used w/24-port manifold to process 12 mL, 20 mL, or 60 mL tubes.

| Description | Qty. | Mfg. Cat. No. |
|------------------------------|------|---------------|
| Long Stem Flow Control Knobs | 6 | 57093 |

96-Well Vacuum Manifolds

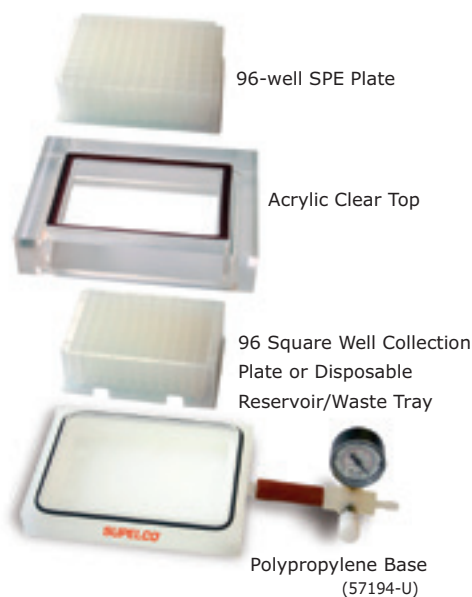
PlatePrep Vacuum Manifold

The PlatePrep vacuum manifold consists of a clear acrylic top allowing for easier inspection of flow rates during SPE 96-well plate processing. The polypropylene base offers excellent chemical resistance while a single remote vacuum gauge/bleed valve controls flow through all the wells.

Use this compact vacuum manifold in conjunction with any of our 96-well plate offerings to process up to 96 samples concurrently. The single valve control, parallel processing capabilities and uniform flow dynamics allow for easier method development, reduce clutter and allow for greater reproducibility. Unused wells can be covered and used at a later date.

Starter Kit (575650-U) Includes:

- A. 1 PlatePrep Vacuum Manifold (57192-U)
- B. 1 96 Sq. Well Collection Plate, 2 mL, PP (575653-U)
- C. 2 Disposable Reservoir/Waste Trays, PVC (575654-U)
- D. 1 96 Sq. Well Pierceable Cap Mat (575655-U)
- E. 5 Reagent Reservoirs (R9259-100EA)
- F. 1 Cluster Tube Rack (CLS4410-960EA)



| Description | Qty. | Mfg. Cat. No. |
|--|------|---------------|
| PlatePrep Vacuum Manifold | 1 | 57192-U |
| 96-Well Plate Starter Kit with PlatePrep Manifold | 1 | 575650-U |
| PlatePrep Vacuum Manifold Replacement Parts | | |
| Gasket/Connector Replacement Kit | 1 | 57195-U |
| Remote Vacuum Gauge/Bleed Valve Assembly | 1 | 57161-U |
| 96-Well SPE Accessory Items | | |
| 96 Sq. Well Collection Plates, 1 mL, PP | 50 | 575652-U |
| 96 Sq. Well Collection Plates, 2 mL, PP | 50 | 575653-U |
| Disposable Reservoir/Waste Tray, PVC | 25 | 575654-U |
| 96 Sq. Well Pierceable Cap Mats | 50 | 575655-U |
| Reagent Reservoirs | 100 | R9259-100EA |
| Cluster Tube Rack | 1 | CLS4410-960EA |

ENVI-Disk™ Accessories

ENVI-Disk™ Holder

Use the ENVI-Disk™ Holder with 47 mm ENVI™-DSK SPE disks (for information on ENVI™-8 and ENVI™-18 DSK SPE disks, see page 19). The unique design of the holder allows each disk to be installed and held firmly in place without wrinkling or tearing. A screw clamp provides uniform pressure on the disk and the sealing surfaces to prevent troublesome leaks – spring-loaded clamps cannot offer the sealing integrity of the ENVI-Disk™ Holder.



Sample Funnel

PTFE Base/Adapter

Flask (order separately)

The unit consists of a 1-liter sample funnel, a threaded screw clamp, a PTFE disk support and a PTFE filter base/adapter with a vacuum attachment fitting. Use 25 x 250 mm test tubes to collect disk eluates. The flask and collection tubes are not included with the holder, but can be purchased separately.

The unit consists of a 1-liter sample funnel, a threaded screw clamp, a PTFE disk support and a PTFE filter base/adapter with a vacuum attachment fitting. Use 25 x 250 mm test tubes to collect disk eluates. The flask and collection tubes are not included with the holder, but can be purchased separately.

| Description | Mfg. Cat. No. |
|--|---------------|
| ENVI-Disk™ Holder | 57173 |
| Flask, 1-liter, 40/35 fitting ¹ | Z290610-1EA |
| Collection Tube, 25 x 250 mm ¹ | 57175 |

¹ Order separately – not included with holder

ENVI-Disk™ Holder Manifold

The ENVI-Disk™ Holder Manifold holds one to six ENVI-Disk™ Holders with flasks, allowing you to simultaneously extract up to six 1-liter samples. Each of the six stations is controlled through an independent flow control valve. These valves are designed to vent the flask to the atmosphere when moved from the open to the closed position. The flow rate is controlled by the needle valve on the manifold.



The unit includes a sturdy polymer base with six stations, six flow control valves, a needle valve, a vacuum gauge and vacuum tubing. A 1-liter glass bottle in the manifold acts as a trap to protect the vacuum source in the event of an overflow from one of the sample flasks.

| Description | Mfg. Cat. No. |
|----------------------------|---------------|
| ENVI-Disk™ Holder Manifold | 57174 |

ENVI-Disk™ Clamp

- Eliminates leaks
- Attaches to any 34/45 tapered flask

When used with a standard 47 mm glass filtration apparatus, the ENVI-Disk™ Clamp creates a better seal, eliminating leaks with SPE extraction disks or when filtering HPLC mobile phase solvents.



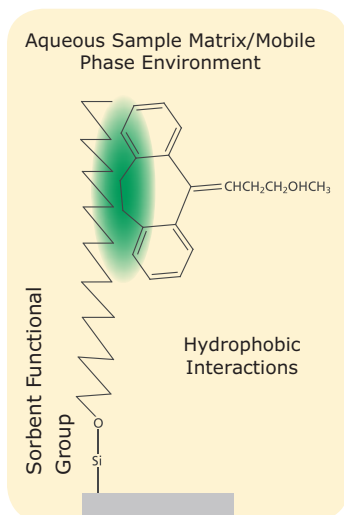
Use only with a filtration glassware funnel base that has a removable filtration stage, such as Supelco® Mobile Phase Filtration Apparatus 1 (58061) or 2 (58062-U), or with a funnel base (58064 or 58068). It cannot be used with a permanent fritted glass filtration stage or stainless steel holder screen.

| Description | Mfg. Cat. No. |
|----------------------------------|---------------|
| ENVI-Disk™ Clamp, 47 mm assembly | 57260-U |
| Replacement PTFE stage | 57261 |

SPE Methodology and Useful Tips

Reversed-Phase SPE

Reversed-phase SPE is considered the least selective retention mechanism when compared to normal-phase or ion-exchange SPE. In other words, it may be difficult for a reversed-phase method or the bonded-chemistry to differentiate between molecules that are structurally similar. However, because reversed-phase will retain most molecules with any hydrophobic character, it is very useful for extracting analytes that are very diverse in structure within the same sample.



Retention Mechanism: Non-polar or hydrophobic interactions

- Van der Waals or dispersion forces

Sample Matrix: Aqueous samples

- Biological fluids (serum, plasma, urine)
- Aqueous extracts of tissues
- Environmental water samples
- Wine, beer and other aqueous food & beverage samples

Analyte Characteristics: Analytes exhibiting non-polar functionalities

- Most organic analytes
- Alkyl, aromatic, alicyclic functional groups

Elution Scheme: Disrupt reversed-phase interaction with solvent or solvent mixtures of adequate non-polar character

- Methanol, acetonitrile, dichloromethane
- Buffer/solvent mixtures

Common Applications

- Drugs and metabolites in biological fluids
- Environmental pollutants in water
- Pesticide and other contaminants in aqueous extracts from tissue & solids

Basic Steps

1. Sample Pre-treatment – For interference laden samples (e.g., biological fluids), dilute samples 1:1 with buffer. pH manipulation may be important when dealing with ionizable compounds. A compound's ionization state can drastically change its retention and elution characteristics on a given SPE sorbent.

When an analyte is in its neutral form, it becomes more hydrophobic and retention is strengthened under reversed-phase conditions. Adjusting the sample pH to 2 pH units above or below the compound's pK_a (depending on the functional group) will effectively neutralize or ionize the compound. When dealing with tissues and other solids, conduct a solid-liquid extraction or homogenization using a buffer. Solvents of non-polar character (including methanol and isopropanol) disrupt interaction between the compound and sorbent functional groups.

To avoid clogging, it may be necessary to centrifuge, dilute and/or pre-filter the sample prior to introducing it to the SPE phase.

2. Conditioning/Equilibration – Conditioning wets or activates the bonded phases to ensure consistent interaction between the analyte and the sorbent functional groups. Reversed-phase sorbents are often conditioned with 1-2 tube volumes of a water miscible solvent such as methanol or acetonitrile.

Equilibration introduces a solution similar to the sample matrix in terms of solvent strength and pH in order to maximize retention. 1-2 tube volumes of buffer (used in sample pre-treatment) or water are good choices for reversed-phase equilibration.

- 3. Sample Load** – Apply sample (from step 1) at a consistent and reduced flow rate of ~1-2 drops/second to ensure optimal interaction time & retention.
- 4. Wash** – Sample interferences are often co-retained with compounds of interest during sample load. A wash step is necessary to elute interferences without prematurely eluting compounds of interest. 5-20% methanol in water or sample pre-treatment buffer are typical for wash solvents.
- 5. Elution** – Disrupt hydrophobic interactions between the analyte and sorbent functional groups with an organic solvent or solvent combination of sufficient non-polar character. Example elution solvents are 1-2 volumes of methanol or acetonitrile.

pH manipulation during elution can often improve recovery when dealing with ionizable compounds. In their ionic form, basic and acidic compounds become more polar, weakening reversed-phase interaction, possibly allowing for weaker elution solvents and/or reduced elution volumes.
- 6. Eluate** – Post-treatment is often necessary to evaporate and reconstitute the SPE eluate in mobile phase prior to LC analysis. GC analysis often requires further SPE eluate concentration and/or possible matrix exchange with a more volatile solvent.

SPE Tips

- Drug-protein binding should be disrupted during sample pre-treatment.
Strategies include:
 - 40 μ L of 2% disodium EDTA per 100 μ L mouse plasma
 - 40 μ L of 2% formic acid per 100 μ L mouse plasma
 - Other possible reagents (per 100 μ L matrix): 40 μ L of 2% TCA, 40 μ L of 2% acetic acid, 40 μ L of 2% TFA, 40 μ L of 2% phosphoric acid, or 200 μ L MeCN (protein ppt.).
 - If the SPE eluate needs to be evaporated prior to analysis, pass vacuum air through the SPE tube for ~10 minutes prior to elution. This will remove residual moisture that may prolong evaporation.
- Consistent and slow flow rate (1-2 drops per second) during sample load and elution will improve recovery and reproducibility.
- Reduce bed weight to minimize elution volume.
- Increase bed weight to retain more polar compounds
- Concern for sorbent overdrying is only critical during methanol conditioning.
- A pre-conditioning solvent such as dichloromethane (or solvent used for elution) can be used before conditioning to remove any impurities on the SPE tube that can interfere with subsequent analysis.

Ion-Exchange and Mixed-Mode SPE

Retention Mechanism: Electrostatic attraction of charged functional groups of the analyte(s) to oppositely charged functional groups on the sorbent. Combination of reversed-phase and ion-exchange for mixed-mode

Sample Matrix: Aqueous or organic samples of low salt concentration (< 0.1 M)

- Biological fluids
- Solution phase synthesis reactions

Analyte Characteristics:

- Use cation-exchange for isolating basic compounds: primary, secondary, tertiary and quarternary amines
- Use anion-exchange for isolating acidic compounds: carboxylic acids, sulphonic acids and phosphates

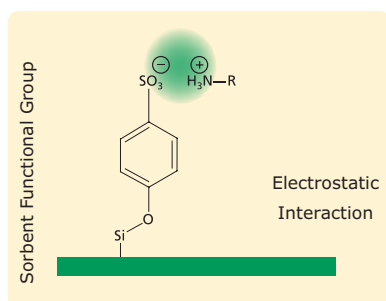
Elution Scheme: Electrostatic interactions disrupted via:

- pH modification to neutralize compound and/or sorbent functional groups
- Increase salt concentration (>1 M); or use a more selective counter-ion to compete for ion-exchange binding sites

Common Applications:

- Drugs of abuse and pharmaceutical compounds in biological fluids
- Fatty acids removal in food/agricultural samples
- Cleanup of synthetic reactions
- Organic acids from urine
- Herbicides in soil

In order for electrostatic retention to occur, both analyte and sorbent functional groups must be in their ionized form. This is done through strict pH control of the sample matrix. For basic analytes, the pH should be adjusted to at least 2 pH units below the molecule's pK_a . For acidic analytes, the pH should be adjusted to at least 2 pH units above the molecule's pK_a .



To elute, the opposite is true. By adjusting the pH of the eluant to at least two pH units above or below the analytes' and/or sorbent's pK_a , one can effectively neutralize one or both functional groups; disrupting the electrostatic interaction allowing for elution to occur.

Note: Because the kinetic exchange processes between sample and sorbent functional groups are considerably slower for ion-exchange than for normal and reversed-phase, flow rates should be drop wise (~1 drop/second). One may also need to increase elution and wash volumes allowing for sufficient residence time for the mobile phase and stationary phase to interact.

Basic Steps

1. Sample Pre-treatment – Salt concentration should be less than 0.1 M. Dilute sample 1:1 with buffer of appropriate pH to ensure analyte functional groups are ionized.

Examples:

- Basic compounds: dilute with 10-25 mM buffer (e.g., potassium phosphate or ammonium acetate), pH 3-6
- Acidic compounds: dilute with 10-50 mM buffer (e.g., acetate buffer), pH 7-9

For interference laden samples (e.g. biological fluids) containing varying levels of salt concentration, use mixed-mode SPE technology.

2. Condition/Equilibration – If samples are in a non-polar solvent, the same solvent should be used to condition the SPE device. For aqueous samples, condition with 1-2 tube volumes of methanol or acetonitrile. Equilibrate with buffer similar/identical in pH and salt concentration to buffer used in the sample pre-treatment.

3. Sample Load – Apply sample (from step 1) at a consistent and reduced flow rate of ~1 drop/second to ensure optimal retention. Mass transfer kinetics of ion-exchange SPE are slower than reversed-phase and normal-phase. Reduced flow rate is critical for consistent recovery.

4. Wash – Adequate control of pH and ionic strength should be maintained to prevent premature elution of the analytes of interest. Use buffer of appropriate pH (e.g. buffer used in sample pre-treatment) to remove polar interferences. More hydrophobic interferences can be removed using up to 100% methanol diluted in sample pre-treatment buffer.

5. Elution – Elute at a consistent and reduced flow rate of ~1 drop/second to ensure optimal compound desorption. The most common elution strategy is by pH manipulation. Also, most ion-exchangers exhibit some mixed-mode behavior. Addition of organic modifier is necessary to disrupt secondary reversed-phase interactions.

Examples:

- Basic compounds: elute with 2-5% ammonium hydroxide in 50-100% methanol
- Acidic compounds: elute with 2-5% acetic acid in 50-100% methanol.

Other elution strategies:

- Use an SPE eluate of higher salt concentration (>1 M)
- Use a more selective counter-ion to compete for ion-exchange binding sites

6. Eluate Post-treatment – A number of elution strategies are available. Various elution strategies should be tested and optimized to minimize eluate post-treatment.

Counter Ion Selectivity and Ion Exchange:

Counter ion selectivity is defined as the degree to which a counter ion is capable of competing with other counter ions for the functional group of an ion exchanger sorbent. Retention is facilitated by having a sorbent and/or sample matrix pre-equilibrated with a counter ion that is less selective than the analyte functional group (minimum competition). Analyte elution is facilitated by using buffers with counter ions more selective than analyte functional group.

For Cation Exchangers:

- $Ca^{2+} > Mg^{2+} > K^+ > Mn^{2+} > RNH_3^+ > NH_4^+ > Na^+ > H^+ > Li^+$

For Anion Exchangers:

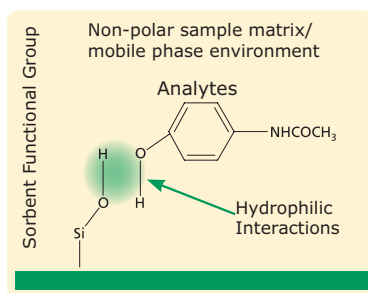
- Benzene Sulphonate > Citrate > $HSO_4^- > NO_3^- > HSO_3^- > NO_2^- > Cl^- > HCO_3^- > HPO_4^- > Formate > Acetate > Propionate > F^- > OH^-$

To change to a higher selective ion, pass 2-5 bed volumes of 1 N solution of the new counter ion through sorbent. To change to a lower selective ion, pass 5-6 bed volumes of 1 N solution of the new counter ion through sorbent.

Note: Number of bed volumes is dependent on how much less selective the new counter ion is than the present one on the sorbent.

Normal-Phase SPE

In order for polar retention to occur between the sorbent and the sample, the analyte must be introduced to the SPE device in a non-polar sample or mobile phase environment. Therefore, typical sample matrices that can be employed in normal-phase SPE include hydrocarbon or fatty oils diluted in an organic solvent, hexane, isooctane, chlorinated solvents, THF, diethyl ether and ethyl acetate.



Most organic analytes exhibit some polar functionalities that can be exploited for normal-phase separation. Because many molecules exhibit polar functionality, each interaction can provide different levels of selectivity offering highly selective separations of compounds very similar in structure.

Retention Mechanism: Polar Interactions

- Hydrogen bonding, pi-pi, dipole-dipole and induced dipole-dipole

Sample Matrix: Non-polar samples

- Organic extracts of solids
- Very non-polar solvents
- Fatty oils, hydrocarbons

Analyte Characteristics: Analytes exhibiting polar functionalities

- Hydroxyl groups, carbonyls, amines, double bonds
- Hetero atoms (O, N, S, P)
- Functional groups with resonance properties

Elution Scheme: Polar interactions disrupted with a more polar solvent or solution

- Acetonitrile, methanol, isopropanol
- Combinations of buffer/solvent or solvent/solvent mixtures

Common Applications:

- Cleanup of organic extracts of soils and sludge
- Fractionation of petroleum hydrocarbons
- PCBs in transformer oil
- Isolation of compounds in cosmetics

Basic Steps

- 1. Sample Pre-treatment** – Liquid samples should be initially extracted or diluted with a non-polar solvent such as hexane or a chlorinated solvent. Soil, sediment and other solid samples are initially extracted (soxhlet or sonication) with a non-polar solvent, and concentrated prior to SPE cleanup. Aqueous residues in the sample can reduce normal-phase retention. It may be necessary to further dry the organic extract with sodium sulfate or magnesium sulfate prior to SPE.
- 2. Condition/Equilibration** – Condition and equilibrate with 2-3 tube volumes of a non-polar solvent similar or identical to sample matrix resulting from sample pre-treatment.

3. Sample Load – Apply sample (from step 1) at a consistent and reduced flow rate of ~1-2 drops/second to ensure optimal retention. The compounds should be in a non-polar solvent (e.g., hexane) for optimal retention. Note that methanol and acetonitrile are often used as elution solvents in normal-phase SPE and will often not promote compound retention during sample load.

4. Wash – Sample interferences are often co-retained with compounds of interest during sample load. A wash step is necessary to elute interferences without prematurely eluting compounds of interest. In normal-phase SPE, 1-2 tube volumes of solvent used in sample pre-treatment and conditioning can be used during wash.

5. Elution – Disrupt polar interactions with a solvent or solvent/buffer mixture more polar than both the sample and wash solutions. Typical elution solvents include water miscible organic solvents such as acetone, acetonitrile, methanol and isopropanol. Eluting with increasingly polar solvents or solvent mixtures in succession can also fractionate multiple compound classes. See “Common Normal-Phase Solvents” table for assistance.

6. Eluate Post-treatment – Normal-phase SPE is often followed by GC analysis, and therefore requires a volatile sample matrix prior to injection. Use sodium sulfate or magnesium sample to remove residual moisture. Further SPE eluate concentration may also be necessary prior to analysis.

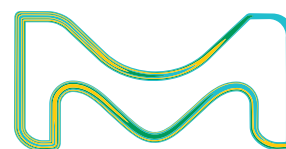
Common Normal-Phase Solvents

| Solvent | Elutropic (e°) or Elution Strength on Silica | |
|--------------------------------------|--|---------------------------------|
| Hexane | 0.00 | Promotes Normal-Phase Retention |
| Isooctane | 0.00 | |
| Carbon tetrachloride | 0.14 | |
| Toluene | 0.22 | |
| Benzene | 0.27 | |
| <i>tert</i> -Butyl methyl ether | 0.29 | |
| Chloroform | 0.31 | |
| Methylene chloride (dichloromethane) | 0.32 | |
| Diethyl ether | 0.29 | |
| Ethyl acetate | 0.43 | |
| Tetrahydrofuran | 0.35 | Promotes Normal-Phase Elution |
| Acetone | 0.45 | |
| Acetonitrile | 0.50 | |
| 40% methanol in acetonitrile | 0.67 | |
| 20% methanol in diethyl ether | 0.65 | |
| 20% methanol in methylene chloride | 0.63 | |
| Isopropanol | 0.63 | |
| Methanol | 0.73 | |
| Water | >0.73 | |
| Acetic acid | >0.73 | |

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