The Complete Guide to Solid Phase Extraction (SPE)

A method development and application guide
Solid Phase Extraction (SPE) is a very targeted form of sample preparation that allows you to isolate your analyte of interest while removing any interfering compounds that may be in your sample.

- Ultra clean extracts
- Concentration of samples for better chromatographic results
- Solvent switching for GC or LC compatibility
- Longer column lifetime and improved chromatographic results

www.phenomenex.com/SPE

If Strata®-X or Strata SPE products do not perform as well or better than your current SPE product of similar phase, mass and size, return the product with comparative data within 45 days for a FULL REFUND.

Getting Started
Follow 3 easy steps and start implementing your complete SPE method.

Select a Sorbent (Step 1) ........................................ p. 4-6

Sample Pre-treatment (Step 2) .................................. p. 7

General Starting Methods
for Strata®-X (Step 3a) .......................................... pp. 8-11
for Strata (Step 3b) ............................................... pp. 12-17

Industry Applications ........................................... pp. 18-24

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Selecting The Right Sorbent: Strata® Silica-Based and Strata-X Polymer-Based Sorbents

Identify the SPE Retention Mechanism

<table>
<thead>
<tr>
<th>Sample Matrix</th>
<th>Aqueous</th>
<th>Organic</th>
</tr>
</thead>
<tbody>
<tr>
<td>Water, Urine, Plasma, Blood, Tissue Prep.</td>
<td>Add H2O</td>
<td>Add Hexane</td>
</tr>
<tr>
<td></td>
<td>Not water miscible, Hexane, Dichromethane</td>
<td></td>
</tr>
</tbody>
</table>

Suggested Retention Mechanism

- **Reversed Phase**: Strata-X, XL, C18, Phenyl, C8
- **Ion-Exchange**: Strata-X, C18-E, C8, X-A, XL-CW, XL-A, X-AW, SAX, SCX, WAX, NH2, ABW, Screen-A, Screen-C
- **Normal Phase**: Strata NH2, CN, Si-1, FL-PR, EPH, AL-N

Determine the Sorbent Chemistry

<table>
<thead>
<tr>
<th>SPE Mechanism</th>
<th>Analyte Functional Group</th>
<th>Sorbent Functional Group</th>
<th>Strata-X Sorbent</th>
<th>Strata Sorbent</th>
</tr>
</thead>
<tbody>
<tr>
<td>Reversed Phase</td>
<td>hydrocarbon, aromatic</td>
<td>C18-E, C18-U, C18-T</td>
<td>Strata-X-C</td>
<td>Strata-C</td>
</tr>
<tr>
<td>Normal Phase</td>
<td>OH, hydroxyl, NH2, amino</td>
<td>CN, NH2, Si-1, CN, EPH</td>
<td>Strata-X-CW</td>
<td>Strata-CW</td>
</tr>
</tbody>
</table>

**Sorbent Properties**

- **Table 1a. SPE Overview**
  - Increase Detection Sensitivity by removing matrix contaminants
  - Increase Column Lifetime by removing matrix contaminants
  - Quality Guaranteed by more than 20 QA and QC measures
  - Increase Reproducibility with robust methods
  - Save Time by processing multiple samples simultaneously or automating method
  - Specific Selectivity for your target analytes
  - Decreased Solvent Consumption with the highest loadability
  - Decreased Blow-down Time with smaller elution volumes
  - Decreased Sample Variation with deconditioning resistant sorbent
  - pH Stable from 1-14

- **Table 2a. Select Your Particle and Pore Size**
  - **Strata-X**
    - Strata-X, X-C, X-A, X-CW, X-AW
    - Particle & Pore Size: 33 μm, 85 Å
    - Suggested Retention Mechanism:
      - High Concentration Samples: 4 – 16 bed volumes
      - Small Target Analytes (< 10 kDa)
      - Large Target Analytes (> 10 kDa)
      - Large Volume Samples
      - Viscous Samples
  - **Strata**
    - Strata-XL, XL-C, XL-CW, XL-A, XL-AW
    - Particle & Pore Size: 100 μm, 300 Å

- **Table 3a. Polymer-Based Sorbents Loading Capacities**
  - **Blood, serum, plasma**
    - Sorbent Mass: 30 mg
    - Strata-X: 250 µL, 125 µL
    - Strata: 1 mL, 500 µL
  - **Urine**
    - Sorbent Mass: 30 mg
    - Strata-X: 1 mL
    - Strata: 500 µL
  - **Filtered tissue homogenates**
    - Sorbent Mass: 60 mg
    - Strata-X: 100 mg
    - Strata: 50 mg
  - **Water (particulate-free)**
    - Sorbent Mass: 200 mg
    - Strata-X: 100 – 400 mL
    - Strata: 50 – 200 mL
  - **Water (particulate-laden)**
    - Sorbent Mass: 500 mg
    - Strata-X: 100 – 400 mL
    - Strata: 50 – 200 mL

- **Table 4a. Sorbent Wash and Elution Volumes**
  - The volume of solvent needed for the wash and elution steps is directly related to the mass of sorbent in the SPE tube and more specifically the "bed volume" of the SPE device. Typically 4 – 10 bed volumes are used in SPE methods.

**Strata-X Polymeric SPE**

- **Step 1. Select a Sorbent**
- **Step 1a. Strata-X Polymeric SPE**
- **Step 2. Determine the Sorbent Chemistry**
- **Step 3. Determine the Sorbent Properties**
- **Step 4. Determine the Sorbent Composition**
- **Step 5. Strata-X Polymeric SPE**
- **Step 6. Strata-X Polymeric SPE**
- **Step 7. Strata-X Polymeric SPE**
Step 1b

Strata® Silica-Based SPE

Sorbent Properties

Table 1b. SPE Overview

<table>
<thead>
<tr>
<th>Strata</th>
<th>Strata-X</th>
</tr>
</thead>
<tbody>
<tr>
<td>Increase Detection Sensitivity by removing matrix contaminants</td>
<td>*</td>
</tr>
<tr>
<td>Increase Column Lifetime by removing matrix contaminants</td>
<td>*</td>
</tr>
<tr>
<td>Quality Guaranteed by more than 20 QA and QC measures</td>
<td>*</td>
</tr>
<tr>
<td>Increase Reproducibility with robust methods</td>
<td>*</td>
</tr>
<tr>
<td>Save Time by processing multiple samples simultaneously or automating method</td>
<td>*</td>
</tr>
<tr>
<td>Specific Selectivity for your target analytes</td>
<td>*</td>
</tr>
<tr>
<td>Decreased Solvent Consumption with the highest loadability</td>
<td>*</td>
</tr>
<tr>
<td>Decreased Blow-down Time with smaller elution volumes</td>
<td>*</td>
</tr>
<tr>
<td>Decreased Sample Variation with deconditioning resistant sorbent</td>
<td>*</td>
</tr>
<tr>
<td>pH Stable from 1-14</td>
<td></td>
</tr>
</tbody>
</table>

Table 2b. Silica-Based Sorbents Loading Capacities

<table>
<thead>
<tr>
<th>Sample Matrix</th>
<th>Sorbent Mass</th>
</tr>
</thead>
<tbody>
<tr>
<td>Blood, serum, plasma</td>
<td>50 mg sorbent per 250 µL</td>
</tr>
<tr>
<td>Urine</td>
<td>50 mg sorbent per 500 µL</td>
</tr>
<tr>
<td>Filtered tissue homogenates</td>
<td>100 mg sorbent per 100 mg tissue</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Environmental Samples</th>
<th>Sorbent Mass</th>
</tr>
</thead>
<tbody>
<tr>
<td>Water (particulate-free) drinking</td>
<td>500 mg/100 mL - 500 mL sample</td>
</tr>
<tr>
<td>Water (particulate-laden) rivers, runoff, etc.</td>
<td>1 g/100 mL - 500 mL sample</td>
</tr>
<tr>
<td>Soil extracts</td>
<td>1 g/100 g of soil extract</td>
</tr>
</tbody>
</table>

Table 3b. Sorbent Wash and Elution Volumes*

The volume of solvent needed for the wash and elution steps is directly related to the mass of sorbent in the SPE tube and more specifically the "bed volume" of the SPE device. Typically 4 - 16 bed volumes are used in SPE methods.

<table>
<thead>
<tr>
<th>Sorbent Mass</th>
<th>Practical Minimum Wash and Elution Volume 4 bed volumes</th>
<th>Recommended Wash and Elution Volume 8 bed volumes</th>
</tr>
</thead>
<tbody>
<tr>
<td>10 mg</td>
<td>60 µL, 300 µL, 600 µL, 900 µL, 1.2 mL, 3 mL, 6 mL, 12 mL, 30 mL, 60 mL</td>
<td>120 µL, 600 µL, 1.2 mL, 1.8 mL, 2.4 mL, 6 mL, 12 mL, 24 mL, 60 mL, 120 mL</td>
</tr>
<tr>
<td>50 mg</td>
<td>300 µL, 600 µL, 900 µL, 1.2 mL, 3 mL, 6 mL, 12 mL, 30 mL, 60 mL</td>
<td></td>
</tr>
<tr>
<td>100 mg</td>
<td>600 µL, 900 µL, 1.2 mL, 3 mL, 6 mL, 12 mL, 30 mL, 60 mL</td>
<td></td>
</tr>
<tr>
<td>200 mg</td>
<td>900 µL, 1.2 mL, 3 mL, 6 mL, 12 mL, 30 mL, 60 mL</td>
<td></td>
</tr>
<tr>
<td>500 mg</td>
<td>1.2 mL, 3 mL, 6 mL, 12 mL, 30 mL, 60 mL</td>
<td></td>
</tr>
<tr>
<td>1 g</td>
<td>3 mL, 6 mL, 12 mL, 30 mL, 60 mL</td>
<td></td>
</tr>
<tr>
<td>2 g</td>
<td>6 mL, 12 mL, 30 mL, 60 mL</td>
<td></td>
</tr>
<tr>
<td>5 g</td>
<td>12 mL, 30 mL, 60 mL</td>
<td></td>
</tr>
<tr>
<td>10 g</td>
<td>30 mL, 60 mL</td>
<td></td>
</tr>
</tbody>
</table>

*The elution volumes are specific to the chemical nature of the analyte being extracted. Its concentration in the sample, the chemical nature of the eluting solvent and the bed mass used. The above is a guideline. An elution study should be conducted to determine the appropriate volume to use.

Strata-X

Step 2

Sample Pre-treatment

Reproducible, high efficiency solid phase extraction requires that the sample be made liquid prior to loading onto a SPE device. The SPE sample should meet the following conditions:

- Liquid of low viscosity (to pass through the cartridge)
- Low solids or particulate contaminants (to prevent clogging)
- Solvent composition that is suitable for retention (each mechanism has different matrix solvent composition requirements for proper retention)

Biological Samples (liquid)

| Urea, Whole blood, Serum, Plasma, Bile, etc. | Dilute sample 1:2 with appropriate buffer, precipitate proteins if proteinaceous (ZnSO4, ACN), hydrolyze urinary glucuronides, disruption of protein binding (sonication, enzymatic, acids/bases). |

Biological Samples (solid)

| Organ tissues, Feces, GI contents | Homogenize with organic or aqueous solvent depending upon analyte solubility. Select, decontaminate and filter supernatant; perform direct Matrix Solid Phase Dispersion (MSPD) extraction on tissue. |

Sample Matrix

| Water (waste, river, etc.) | Buffer to appropriate pH and filter particulates from sample. |
| Soil, Sludge | Homogenize with organic or aqueous solvent depending upon analyte solubility. Select, decontaminate and filter supernatant; perform Soxhlet extraction. |
| Ointments, Creams | Oil-based - Dissolve in non-polar organic (hexane) and extract via polar SPE. Water-based - Dissolve in water or water miscible organic (methanol) and extract via non-polar SPE. |
| Fruit, Vegetable, Herbs | Homogenize with organic or aqueous solvent depending upon analyte solubility and filter supernatant. Use appropriate SPE mechanism for the dissolution solvent (hexane = polar mechanism; aqueous = non-polar mechanism; methanol/ACN = either non-polar or polar after proper dilution). |

Support@Phenomenex.com

Dedicated sample preparation team available to assist your method development needs.

Phenomenex | WEB: www.phenomenex.com
**Strata-X Polymeric SPE Phase Overview**

- Clean extracts from biological sample matrices
- Streamlined method development and simple processing

<table>
<thead>
<tr>
<th>Strata-X Phase</th>
<th>Functional Group</th>
<th>Mode</th>
<th>Analyte</th>
<th>Recommended Alternative to Waters®</th>
</tr>
</thead>
<tbody>
<tr>
<td>Strata-X</td>
<td></td>
<td>Reversed Phase</td>
<td>Polar and Non-Polar</td>
<td>Oasis® HLB</td>
</tr>
<tr>
<td>Strata-X-C</td>
<td></td>
<td>Reversed Phase and Strong Cation-Exchange</td>
<td>Bases</td>
<td>Oasis MCX</td>
</tr>
<tr>
<td>Strata-X-CW</td>
<td></td>
<td>Reversed Phase and Weak Cation-Exchange</td>
<td>Bases (including Quaternary Amines)</td>
<td>Oasis WCX</td>
</tr>
<tr>
<td>Strata-X-A</td>
<td></td>
<td>Reversed Phase and Strong Anion-Exchange</td>
<td>Acids</td>
<td>Oasis MAX</td>
</tr>
<tr>
<td>Strata-X-AW</td>
<td></td>
<td>Reversed Phase and Strong Anion-Exchange</td>
<td>Acids (including Sulfonic acids)</td>
<td>Oasis MAX</td>
</tr>
<tr>
<td>Strata-XL</td>
<td></td>
<td>Large Particle Reversed Phase</td>
<td>Polar and Non-Polar</td>
<td>Oasis WAX</td>
</tr>
<tr>
<td>Strata-XL-C</td>
<td></td>
<td>Large Particle Reversed Phase and Strong Cation-Exchange</td>
<td>Bases</td>
<td>Oasis MCX</td>
</tr>
<tr>
<td>Strata-XL-CW</td>
<td></td>
<td>Large Particle Reversed Phase and Weak Cation-Exchange</td>
<td>Bases (including Quaternary Amines)</td>
<td>Oasis WCX</td>
</tr>
<tr>
<td>Strata-XL-A</td>
<td></td>
<td>Large Particle Reversed Phase and Strong Anion-Exchange</td>
<td>Acids</td>
<td>Oasis MAX</td>
</tr>
<tr>
<td>Strata-XL-AW</td>
<td></td>
<td>Large Particle Reversed Phase and Weak Anion-Exchange</td>
<td>Acids (including Sulfonic acids)</td>
<td>Oasis WAX</td>
</tr>
</tbody>
</table>

**Strata-X / Strata-XL Reversed Phase**

For Neutral Compounds

<table>
<thead>
<tr>
<th>Condition</th>
<th>Equilibrate 1 mL Methanol</th>
</tr>
</thead>
<tbody>
<tr>
<td>Load</td>
<td>Diluted Sample</td>
</tr>
<tr>
<td>Wash</td>
<td>1 mL 5-60 % Methanol</td>
</tr>
<tr>
<td>Elute</td>
<td>2x 500 µL 2 % Formic Acid in Methanol/Acetonitrile</td>
</tr>
</tbody>
</table>

**SPE Method Development Tool**

Develop SPE methods for sample cleanup and concentration in under one minute. [www.phenomenex.com/mdtool](http://www.phenomenex.com/mdtool)

**Search Hundreds of Applications**

Know the name of your analyte? Immediately find key Sample Prep applications for small molecules and biomolecules by entering the name or the chemical properties of the analyte. [www.phenomenex.com/applications](http://www.phenomenex.com/applications)

*Based on 30 mg/1 mL sorbent mass. The above is a convenient starting point for SPE method development. Further optimization may be required to tailor the method to your specific needs.*
**Step 3a  General Starting Methods: Strata®-X (cont’d)**

**Strata-X-C / Strata-XL-C**

**Strong Cation-Exchange & Reversed Phase**

For Bases with $pK_a \leq 10.5$

- **Condition**
  - 1 mL Methanol
- **Equilibrate**
  - 1 mL Acidified Water
- **Load**
  - Diluted Acidified Sample
- **Wash 1**
  - 1 mL 0.1 N HCl in Water (collect this fraction to analyze Polar Neutrals)
- **Wash 2**
  - 1 mL 0.1 N HCl in Methanol (collect this fraction to analyze Neutrals/Acids)
- **Elute Bases**
  - 2x 500 µL 5% NH₄OH in Methanol

**Strong Cation-Exchange:
 sulfonic acid ligand**

**Strata-X-CW / Strata-XL-CW**

**Weak Cation-Exchange & Reversed Phase**

For Bases with $pK_a > 8$

- **Condition**
  - 1 mL Methanol
- **Equilibrate**
  - 1 mL Water, pH 6-7
- **Load**
  - Diluted Sample, pH 6-7
- **Wash 1**
  - 1 mL 25 mM Ammonium Acetate Buffered, pH 6-7
- **Wash 2**
  - 1 mL Methanol (collect this fraction to analyze Neutral/Bases)
- **Elute Any Base**
  - 2x 500 µL 5% Formic Acid in Methanol

**Weak Cation-Exchange:
carboxylic acid ligand**

**Strata-X-A / Strata-XL-A**

**Strong Anion-Exchange & Reversed Phase**

For Acids with $pK_a > 2$

- **Condition**
  - 1 mL Methanol
- **Equilibrate**
  - 1 mL Water, pH 6-7
- **Load**
  - Diluted Sample, pH 6-7
- **Wash 1**
  - 1 mL 25 mM Ammonium Acetate Buffered, pH 6-7
- **Wash 2**
  - 1 mL Methanol (collect this fraction to analyze Neutral/Bases)
- **Elute Weak Acids**
  - 2x 500 µL 5% Formic Acid in Methanol

**Weak Anion-Exchange:
di-amino ligand**

**Strata-X-AW / Strata-XL-AW**

**Weak Anion-Exchange & Reversed Phase**

For Acids with $pK_a \leq 5$

- **Condition**
  - 1 mL Methanol
- **Equilibrate**
  - 1 mL Water, pH 6-7
- **Load**
  - Diluted Sample, pH 6-7
- **Wash 1**
  - 1 mL 25 mM Ammonium Acetate Buffered, pH 6-7
- **Wash 2**
  - 1 mL Methanol
- **Elute Any Acid**
  - 2x 500 µL 5% NH₄OH in Methanol
- **Elute Weak Acids**
  - 2x 500 µL 5% Formic Acid in Methanol

**Weak Anion-Exchange:
di-methylbutyl quaternary amine ligand**

*Based on 30 mg/1 mL sorbent mass. The above is a convenient starting point for SPE method development. Further optimization may be required to tailor the method to your specific needs.
# Phenomenex Normal Phase Sorbents

<table>
<thead>
<tr>
<th>Sorbent</th>
<th>Method</th>
<th>Waters® Sep-Pak®</th>
<th>Agilent® SampliQ®</th>
<th>Biotage® ISOLUTE®</th>
<th>UCT® CleanScreen®</th>
<th>Supelco® Discovery®</th>
</tr>
</thead>
<tbody>
<tr>
<td>Si-1 (Silica)</td>
<td>C18</td>
<td>Sample EC</td>
<td>Bond Elut C18</td>
<td>C18</td>
<td>C18</td>
<td>DSC-18</td>
</tr>
<tr>
<td>FL-PR (Florisil®)</td>
<td>C18</td>
<td>Sample EC</td>
<td>Bond Elut C18</td>
<td>C18</td>
<td>C18</td>
<td>DSC-18</td>
</tr>
</tbody>
</table>

# Phenomenex Reversed Phase Sorbents

<table>
<thead>
<tr>
<th>Sorbent</th>
<th>Method</th>
<th>Waters® Sep-Pak®</th>
<th>Agilent® SampliQ®</th>
<th>Biotage® ISOLUTE®</th>
<th>UCT® CleanScreen®</th>
<th>Supelco® Discovery®</th>
</tr>
</thead>
<tbody>
<tr>
<td>CN</td>
<td>C8</td>
<td>Sample EC</td>
<td>Bond Elut C8</td>
<td>C8(EC)</td>
<td>C8</td>
<td>DSC-8</td>
</tr>
<tr>
<td>SDB-L</td>
<td>C8</td>
<td>Sample EC</td>
<td>Bond Elut LMS</td>
<td>101</td>
<td>StyleScreen® DVB</td>
<td>DSC-PS/DVB</td>
</tr>
</tbody>
</table>

**Recommended Alternative to:**
- Waters® Sep-Pak®
- Agilent® SampliQ®
- Biotage® ISOLUTE®
- UCT® CleanScreen®
- Supelco® Discovery®

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### Step 3b General Starting Methods: Strata®

**Strata Silica-Based SPE Phase Overview**

<table>
<thead>
<tr>
<th>Strata Phase</th>
<th>Phase Benefits</th>
<th>Recommended Method (See pp. 16-17)</th>
<th>Recommended Alternative to:</th>
</tr>
</thead>
<tbody>
<tr>
<td>C18-E</td>
<td>Extraction of hydrophobic molecules</td>
<td>METHOD 1</td>
<td>Sample C18EC Bond Elut C18</td>
</tr>
<tr>
<td>C18-U</td>
<td>Enhanced cleanup of hydrophobic compounds that contain hydroxy or amine functional groups</td>
<td>METHOD 1</td>
<td>Bond Elut C18-OH C18</td>
</tr>
<tr>
<td>C18-T</td>
<td>Wide pore for the extraction of large hydrophobic molecules (up to 75 kDa)</td>
<td>METHOD 1</td>
<td>Bond Elut C18-EWP</td>
</tr>
<tr>
<td>C8</td>
<td>Extraction of extremely hydrophobic compounds that are retained too tightly on C18-E</td>
<td>METHOD 1</td>
<td>Sample C8 Octyl Bond Elut C8</td>
</tr>
<tr>
<td>Phenyl (PH)</td>
<td>Extraction of aromatic compounds</td>
<td>METHOD 1</td>
<td>Sample Phenyl Bond Elut PH</td>
</tr>
<tr>
<td>CN</td>
<td>Extraction of polar compounds</td>
<td>METHOD 1</td>
<td>Sample Cyano (CN) Bond Elut Cyano-CN (CN-E)</td>
</tr>
<tr>
<td>SDB-L</td>
<td>Extraction of non-polar and polar compounds; pH-resistant sorbent</td>
<td>METHOD 1</td>
<td>Sample DVB Bond Elut ENV Bond Elut LMS</td>
</tr>
</tbody>
</table>

**Recommended Alternative to:**
- Waters® Sep-Pak®
- Agilent® SampliQ®
- Biotage® ISOLUTE®
- UCT® CleanScreen®
- Supelco® Discovery®

---

**Normal Phase Sorbents**

<table>
<thead>
<tr>
<th>Sorbent</th>
<th>Method</th>
<th>Waters® Sep-Pak®</th>
<th>Agilent® SampliQ®</th>
<th>Biotage® ISOLUTE®</th>
<th>UCT® CleanScreen®</th>
<th>Supelco® Discovery®</th>
</tr>
</thead>
<tbody>
<tr>
<td>Si-1 (Silica)</td>
<td>C18</td>
<td>Sample Si</td>
<td>Bond Elut Si</td>
<td>Si</td>
<td>Si</td>
<td>DSC-Si</td>
</tr>
<tr>
<td>FL-PR (Florisil®)</td>
<td>C18</td>
<td>Sample Si</td>
<td>Bond Elut Florisil®</td>
<td>FL</td>
<td>Florisil® PR ENVI-Florisil®</td>
<td></td>
</tr>
<tr>
<td>NH₃</td>
<td>C18</td>
<td>Sample Amino (NH₃)</td>
<td>Bond Elut Aminopropyl (NH₃)</td>
<td>NH₃</td>
<td>Amino Propyl DSC-NH₃</td>
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</tr>
<tr>
<td>CN</td>
<td>C18</td>
<td>Sample Cyano (CN)</td>
<td>Bond Elut Cyano (CN-E)</td>
<td>CN</td>
<td>CN</td>
<td>DSC-CN</td>
</tr>
</tbody>
</table>
### Strata Silica-Based Phase Overview

#### Ion-Exchange Sorbents

<table>
<thead>
<tr>
<th>Strata Phase</th>
<th>Phase Benefits</th>
<th>Sorbent Chemistry</th>
<th>Recommended Method (See pp. 16-17)</th>
<th>Recommended Alternative to:</th>
</tr>
</thead>
<tbody>
<tr>
<td>ABW</td>
<td>Fractionation of neutral compounds such as amides from acidic and basic analytes</td>
<td><img src="image1" alt="ABW Structure" /></td>
<td><img src="image2" alt="Schematic" /></td>
<td>Inspire</td>
</tr>
<tr>
<td>SAX</td>
<td>Extraction of weak anions</td>
<td><img src="image3" alt="SAX Structure" /></td>
<td><img src="image4" alt="METHOD 5" /></td>
<td>Accell Plus QMA Bond Elut SAX Sax Quaternary Amine DSC-SAX</td>
</tr>
<tr>
<td>SCX</td>
<td>Extraction of 1°, 2°, and 3° amines</td>
<td><img src="image5" alt="SCX Structure" /></td>
<td><img src="image6" alt="METHOD 3" /></td>
<td>SampliQ Si-SCX Bond Elut SCX SCX-3 Benzeno Sulfonic Acid DSC-SCX</td>
</tr>
<tr>
<td>WCX</td>
<td>Extraction of quaternary amines</td>
<td><img src="image7" alt="WCX Structure" /></td>
<td><img src="image8" alt="METHOD 5" /></td>
<td>Accell Plus CM Bond Elut CBA CBA Carboxylic Acid DSC-WCX</td>
</tr>
<tr>
<td>Screen-C</td>
<td>Mixed-mode cation-exchange that also provides hydrophobic retention</td>
<td><img src="image9" alt="Screen-C Structure" /></td>
<td><img src="image10" alt="METHOD 3" /></td>
<td>SampliQ C8/Si-SCX Mixed Mode Bond Elut Certify®* HAX Clean Screen® DAU</td>
</tr>
<tr>
<td>Screen-C GF</td>
<td>Large particle size, mixed-mode cation-exchange that also provides hydrophobic retention</td>
<td><img src="image11" alt="Screen-C GF Structure" /></td>
<td><img src="image12" alt="METHOD 4" /></td>
<td>Bond Elut Certify® I HF Xtract® DAU</td>
</tr>
<tr>
<td>Screen-A</td>
<td>Mixed-mode anion-exchange that also provides hydrophobic retention</td>
<td><img src="image13" alt="Screen-A Structure" /></td>
<td><img src="image14" alt="METHOD 5" /></td>
<td>Bond Elut Certify® II HAX Clean Screen THC</td>
</tr>
<tr>
<td>NH₂</td>
<td>Extraction of strong anions</td>
<td><img src="image15" alt="NH₂ Structure" /></td>
<td><img src="image16" alt="METHOD 4" /></td>
<td>NH₂ SampliQ Amino (NH₂) Bond Elut Aminopropyl (NH₂) NH₂ Amino Propyl DSC-NH₂</td>
</tr>
</tbody>
</table>

#### Special Sorbents

<table>
<thead>
<tr>
<th>Sorbent</th>
<th>Description</th>
<th>Method</th>
<th>Alternative to:</th>
</tr>
</thead>
<tbody>
<tr>
<td>Alumina-N (AL-N)</td>
<td>Extraction of polar compounds from food and environmental samples</td>
<td><img src="image17" alt="METHOD 6" /></td>
<td>Alumina-N</td>
</tr>
<tr>
<td>EPH</td>
<td>Fractionation of aliphatic and aromatic hydrocarbons from environmental samples</td>
<td><img src="image18" alt="METHOD 6" /></td>
<td></td>
</tr>
</tbody>
</table>

---

For more detailed information, refer to pages 16-17.
**General Starting Methods: Strata® (cont’d)**

### Method 1
**Condition**
1 mL Methanol

**Equilibrate**
1 mL DI Water

**Load**
Pretreated sample

**Wash**
1 mL 5% Methanol in DI Water, dry under vacuum for 2-5 min

**Elute**
1 mL Methanol

### Method 2
**Condition**
1 mL Methanol

**Equilibrate**
1 mL DI Water, pH 6-7

**Load**
Pretreated sample, pH 6-7

**Wash 1**
1 mL Water, pH 6-7

**Wash 2**
1 mL Methanol, dry under vacuum for 2-5 min

**Elute Any Base**
1 mL 5% Formic Acid in Methanol

**Elute Weak Bases**
1 mL 5% NH4OH in Methanol

### Method 3
**Condition**
1 mL Methanol

**Equilibrate**
1 mL Acidified Water

**Load**
Pretreated sample (acidified)

**Wash 1**
1 mL 0.1N HCl in Water

**Wash 2**
1 mL 0.1N HCl in Methanol, dry under vacuum for 2-5 min

**Elute**
1 mL 5% NH4OH in Methanol

### Method 4
**Condition**
1 mL Methanol

**Equilibrate**
1 mL Water, pH 6-7

**Load**
Pretreated sample, pH 6-7

**Wash 1**
1 mL 25 mM Ammonium Acetate Buffer, pH 6-7

**Wash 2**
1 mL Methanol, dry under vacuum for 2-5 min

**Elute Any Acid**
1 mL 5% NH4OH in Methanol

**Elute Weak Acids**
1 mL 5% Formic Acid in Methanol

### Method 5
**Condition**
1 mL Methanol

**Equilibrate**
1 mL Water

**Load**
Pretreated sample, pH 6-7

**Wash 1**
1 mL 25 mM Ammonium Acetate Buffer, pH 6-7

**Wash 2**
1 mL Methanol, dry under vacuum for 2-5 min

**Elute**
1 mL 5% Formic Acid in Methanol

### Method 6
**Condition**
IPA / DCM

**Equilibrate**
Hexane

**Load**
Pretreated sample

**Wash**
5% DCM in Hexane

**Elute**
1:1 Hexane / DCM or 1:1 Hexane / IPA
Preventing Analyte Loss by Skipping the Dry Down Step using Microelution SPE

Many target analytes, such as peptides and thermolabile compounds, can be lost during dry down steps. To prevent analyte loss and skip the dry down, without losing sensitivity using Strata®-X microelution plates. A new format that provides increased sensitivity for analytes of interest.

SPE Protocol

<table>
<thead>
<tr>
<th>Part No.</th>
<th>Condition</th>
<th>Load</th>
<th>Wash 1</th>
<th>Wash 2</th>
<th>Elute</th>
<th>Dry Down</th>
<th>Inject</th>
</tr>
</thead>
<tbody>
<tr>
<td>8E-S100-AGB</td>
<td>400 µL Methanol</td>
<td>400 µL diluted serum</td>
<td>400 µL diluted serum</td>
<td>240 µL Trifluoroacetic acid/acetonitrile/water (1:74:25)</td>
<td>NOT REQUIRED</td>
<td>10 µL</td>
<td></td>
</tr>
<tr>
<td>8M-S100-4GA</td>
<td>200 µL Methanol</td>
<td>200 µL diluted 4 % Phosphoric acid in water</td>
<td>200 µL diluted 4 % Phosphoric acid in water</td>
<td>2x 35-µL Trifluoroacetic acid/acetonitrile/water (1:74:25)</td>
<td>Dry down under a gentle stream of Nitrogen and reconstitute in 50 µL Trifluoroacetic acid/ acetonitrile/water (1:74:25)</td>
<td>10 µL</td>
<td></td>
</tr>
</tbody>
</table>

**DALDA C8 (peptide) Extracted from Serum**

**Chromatogram after SPE Extraction from a Plasma Matrix**

To learn more about this method and others, visit: www.phenomenex.com/SPE
Amphetamines from Urine Using Microelution SPE

An extraction method to isolate five amphetamines from urine using Strata-X-C Microelution 96-well SPE plates followed by LC/MS/MS analysis. By utilizing the microelution SPE format, the dry down step was skipped saving at least 30 minutes without negatively impacting the sensitivity of our analysis. The five amphetamines were accurately quantified at detection levels down to 25% below the cutoff levels specified by the Substance Abuse and Mental Health Services Administration (SAMHSA).

<table>
<thead>
<tr>
<th>Amphetamine</th>
<th>Concentration (ng/mL)</th>
<th>RT (min)</th>
<th>% Absolute Recovery</th>
<th>% CV (N=8)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Amphetamine</td>
<td>125</td>
<td>1.83</td>
<td>82</td>
<td>13.1</td>
</tr>
<tr>
<td>Methamphetamine</td>
<td>125</td>
<td>2.12</td>
<td>107</td>
<td>15.1</td>
</tr>
<tr>
<td>MDA</td>
<td>62.25</td>
<td>2.15</td>
<td>106</td>
<td>4.2</td>
</tr>
<tr>
<td>MDMA</td>
<td>62.25</td>
<td>2.36</td>
<td>99</td>
<td>15.7</td>
</tr>
<tr>
<td>MDEA</td>
<td>62.25</td>
<td>2.53</td>
<td>108</td>
<td>10.5</td>
</tr>
</tbody>
</table>

Chromatogram of Amphetamines Extracted from Human Urine

Urinary Steroids using Strata-X SPE

We evaluated a variety of silica-based and polymer-based SPE sorbents for the quantification of cortisol, cortisone, prednisolone, and prednisone, each of which provides a different retention mechanism. The evaluation showed that the Strata-X polymer-based SPE sorbent, with a unique elution solvent has been found to be a robust, reproducible, and cost effective sample preparation solution for the laboratory in human urine for all four corticosteroids.

<table>
<thead>
<tr>
<th>Wash Solvent Optimization</th>
</tr>
</thead>
<tbody>
<tr>
<td>Wash 1</td>
</tr>
<tr>
<td>Load 300 µL</td>
</tr>
<tr>
<td>Wash 2 10 % Methanol in Water</td>
</tr>
<tr>
<td>Elute 2x 500 µL of 2 % Formic Acid in Ethyl acetate/Isopropanol (85:15)</td>
</tr>
</tbody>
</table>

Recovery using SPE Sorbents

Recovery using Strata-X Across Low (QCL, 30 ng/mL) and High (QCH, 1600 ng/mL) QC Concentrations
Chlorinated Pesticides in Poultry Tissue Using Strata® Alumina-N SPE

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

Pretreatment Protocol

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

SPE Protocol

<table>
<thead>
<tr>
<th>Part No.</th>
<th>Strata Alumina-N, 2 g/12 mL</th>
</tr>
</thead>
<tbody>
<tr>
<td>Condition</td>
<td>Methanol/Water (86:14) at 10 mL/min until dry</td>
</tr>
<tr>
<td>Equilibrate</td>
<td>Petroleum ether at full cartridge volume at 10 mL/min</td>
</tr>
<tr>
<td>Load</td>
<td>1 mL Pretreated sample</td>
</tr>
<tr>
<td>Elute</td>
<td>Ethyl Ether/Petroleum Ether (1.5:98.5) at full cartridge volume and collect eluent</td>
</tr>
<tr>
<td>Dry Down</td>
<td>Dry down at ambient temperatures under a stream of nitrogen and evaporate to dryness</td>
</tr>
<tr>
<td>Reconstitute</td>
<td>2 mL Hexane</td>
</tr>
</tbody>
</table>

GC / EDC Analysis of Chlorinated Hydrocarbons

A simple yet effective SPE and cleanup method for phenylbutazone from meat with recovery values > 90 %. Highly specific LC/MS/MS data is generated using a Kinetex core-shell column enabling rapid run times under 5 minutes with excellent precision and accuracy.

Phenylbutazone in Ground Meat using Strata®-X-A SPE

% Recovery of Phenylbutazone from Beef Extract at 5 ppb and 75 ppb (µg/kg) n=4

Phenylbutazone and Phenylbutazone-D10 Chemical Structures

Chromatogram of 10 ppb Phenylbutazone

Chromatogram of 10 ppb Phenylbutazone
Polycyclic Aromatic Hydrocarbons using Strata® PAH as Compared to EPA Method 550.1

Polycyclic aromatic hydrocarbon compounds (PAHs) are effectively extracted from water samples while humic acids, which often interfere with chromatographic separation, are removed from the sample using a SPE sorbent. Strata PAH. It was also found that Strata PAH provides consistent, high recoveries of all 16 Polycyclic aromatic hydrocarbon compounds (PAHs) are effectively extracted from water samples while humic acids from Suwannee River are removed.

**PAH % Recoveries from Tap Water**

<table>
<thead>
<tr>
<th>PAH % Recoveries</th>
<th>25</th>
<th>50</th>
<th>75</th>
<th>90</th>
<th>95</th>
</tr>
</thead>
<tbody>
<tr>
<td>Naphthalene</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Acenaphthylene</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Acenaphthene</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Phenanthrene</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Fluoranthene</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Benzo[a]pyrene</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Indeno[1,2,3-cd]pyrene</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Benzo[b]fluoranthene</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Benzo[k]fluoranthene</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Chrysene</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Benzo[g,h,i]perylene</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
</tbody>
</table>

**Effective Removal of Humic Acids**

- Strata PAH % Recoveries: 100%
- Strata C18 % Recoveries: 75%

**Strata®-X Polymeric SPE Sorbents**

**Strata®-X Polymeric SPE Sorbents**

- **Phase:** 50 mg, 100 mg, 200 mg, 500 mg, 1 g
- **Tubes:** 1 mL (100/box), 3 mL (50/box), 6 mL (30/box)

**Strata® Silica-Based SPE Sorbents**

- **Phase:** 50 mg, 100 mg, 200 mg, 500 mg, 1 g
- **Tubes:** 1 mL (100/box), 3 mL (50/box), 6 mL (30/box)

**Strata Mixed-Mode Sorbents (for drugs of abuse)**

- **Phase:** 50 mg, 100 mg, 200 mg, 500 mg, 1 g
- **Tubes:** 1 mL (100/box), 3 mL (50/box), 6 mL (30/box)

**Accessories For Tubes**

- **Part No.:** AHS-7711
- **Description:** Adapter Caps for 1, 3, and 6 mL SPE tubes, polyethylene, with Luer tip (10/pk)

**SPE Tube Vacuum Manifolds**

- **Part No.:** AHS-6024
- **Description:** SPE 8-Position Vacuum Manifold Set, complete assembly as 8-Position Vacuum Manifold Kit

- **Part No.:** AHS-7024
- **Description:** 16-Position Vacuum Manifold Set, complete assembly as 16-Position Vacuum Manifold Kit

- **Part No.:** AHS-7032
- **Description:** 24-Position Vacuum Manifold Set, complete assembly as 24-Position Vacuum Manifold Kit

**NOTE:**

- Each SPE manifold includes the following: vacuum chamber, vacuum gauge assembly, polycarbonate lid with gasket, male and female storage containers, polypropylene needles, 10 support legs, waste container included with collection manifold.
- Blank sample is included with collection manifold.
- The 12-position Collection Rack Assembly consists of 3 support legs, base plates, small plates, medium plates, large plates, and 12 retaining clips.
- The 16-position Collection Rack Assembly consists of 3 support legs, base plates, small plates, medium plates, large plates, and 12 retaining clips.
- The 24-position Collection Rack Assembly consists of 3 support legs, base plates, small plates, medium plates, large plates, and 12 retaining clips.

**SPE Column**

- **Part No.:** AHS-7151
- **Description:** Polymeric SPE, polyethylene, with Luer tip (10/pk)

**Factors for For Tubes**

- **Part No.:** AHS-7110
- **Description:** Adapter Caps for 1, 3, and 6 mL SPE tubes, polyethylene, with Luer tip (10/pk)

**SPE Tube Vacuum Manifolds**

- **Part No.:** AHS-6024
- **Description:** SPE 8-Position Vacuum Manifold Set, complete assembly as 8-Position Vacuum Manifold Kit

- **Part No.:** AHS-7024
- **Description:** 16-Position Vacuum Manifold Set, complete assembly as 16-Position Vacuum Manifold Kit

- **Part No.:** AHS-7032
- **Description:** 24-Position Vacuum Manifold Set, complete assembly as 24-Position Vacuum Manifold Kit
If Strata-X or Strata SPE products do not perform as well or better than your current SPE product of similar phase, mass and size, return the product with comparative data within 45 days for a FULL REFUND.

Strata®-X Polymeric SPE Sorbents
96-Well Plates (2/Box)

Strata-X-AW
8E-S038-AGB
8E-S038-TGB
8E-S038-UGB

Strata-X-A
8E-S123-AGB
8E-S123-TGB
8E-S123-UGB

Strata-X
8E-S100-AGB
8E-S100-TGB
8E-S100-UGB

Strata-X-C
8E-S029-AGB
8E-S029-TGB
8E-S029-UGB

Strata-XL-AW
– 8E-S051-TGB
–

Strata-XL-A
– 8E-S053-TGB
–

Strata-XL
– 8E-S043-TGB
–

Strata-XL-C
– 8E-S044-TGB
–

Strata-XL-CW
– 8E-S052-TGB
–

Strata Silica-Based SPE Sorbents
96-Well Plates (2/Box)

C18-E
8E-S001-CGB
8E-S001-DGB
8E-S001-EGB

C18-U
— 8E-S002-DGB
8E-S002-EGB

C18-T
8E-S004-CGB
8E-S004-DGB
—

C8
8E-S005-CGB
—
—

Phenyl
8E-S006-CGB
—
8E-S006-EGB

Silica
— 8E-S012-DGB
8E-S012-TGB

NH2
8E-S009-CGB
8E-S009-DGB
8E-S009-EGB

SAX
8E-S008-CGB
8E-S008-DGB
8E-S008-EGB

SCX
8E-S010-CGB
8E-S010-DGB
8E-S010-EGB

WCX
8E-S027-CGB
8E-S027-DGB
—

ZB-MultiRes™-1
ID(mm) df(µm) Temp. Limits °C Part No.
20-Meter
0.18
0.18
-60 to 320/340
7FD-G016-08

30-Meter
0.25
0.25
-60 to 320/340
7HG-G016-11

0.32
0.25
-60 to 320/340
7HM-G016-11

0.32
0.50
-60 to 320/340
7HM-G016-17

0.53
0.50
-60 to 320/340
7HK-G016-17

2.6 μm Minibore Columns (mm) SecurityGuard™ ULTRA Cartridges ‡
Phases
30 x 2.1 50 x 2.1 75 x 2.1 100 x 2.1 150 x 2.1 3/pk

Biphenyl
00A-4622-AN
00B-4622-AN
—
00D-4622-AN
00F-4622-AN
AJ0-9209

XB-C18
00A-4496-AN
00B-4496-AN
00C-4496-AN
00D-4496-AN
00F-4496-AN
AJ0-8782

C8
00A-4497-AN
00B-4497-AN
00C-4497-AN
00D-4497-AN
00F-4497-AN
AJ0-8784

2.6 μm Analytical Columns (mm) SecurityGuard™ ULTRA Cartridges ‡
Phases
30 x 4.6 50 x 4.6 75 x 4.6 100 x 4.6 150 x 4.6 250 x 4.6 3/pk

C18
00A-4462-E0
00B-4462-E0
00C-4462-E0
00D-4462-E0
00F-4462-E0
00G-4462-E0
AJ0-8788

5 μm Minibore Columns (mm) SecurityGuard™ ULTRA Cartridges ‡
Phases
50 x 2.1 100 x 2.1 3/pk

C8
00B-4608-AN
00D-4608-AN
AJ0-8784

5 μm Analytical Columns (mm) SecurityGuard™ ULTRA Cartridges ‡
Phases
50 x 4.6 100 x 4.6 150 x 4.6 250 x 4.6 3/pk

C8
00B-4608-E0
00D-4608-E0
00F-4608-E0
00G-4608-E0
AJ0-8770

‡SecurityGuard ULTRA Cartridges required holder, Part No.: AJ0-9000.
The Complete Guide
to Solid Phase Extraction (SPE)
A Method Development and Application Guide

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info@phenomenex.com

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