Ion exchange SPE optimized for amino acid enrichment

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Introduction
Amino acid enrichment is challenging due to the wide diversity of amino acid structures ranging from acidic, neutral, to basic as well as hydrophilic and hydrophobic functional groups. The challenge is compounded by the complex matrices that you must enrich them from. This diverse range of properties needs to be accounted for during extraction of amino acids. In this protocol we provide a method for extraction of the amino acids from a complex matrix with Thermo Scientific™ SOLAμ™ SCX plates. This method is transferable from the 96-well SOLAμ plates to Thermo Scientific™ SOLA™ SPE cartridges.

Important notes
• Making up the amino acid sample in acidic conditions aids in the interaction of the ion exchange media and the amino acids
• Collect flow through from loading step and reapply to stationary phase to minimize losses
• Collect flow through from loading and wash steps and save for analysis while optimizing SPE method to ensure these fractions are free from analyte(s) of interest
• Select the solid phase extraction (SPE) product that matches the capacity required for your sample
• Use the solvent step volumes appropriate to your SPE bed volume, for example 200 µL for 2 mg SOLAμ and 500 µL for 10 mg SOLA beds

Materials required
• Fisher Chemical Methanol, Optima™ LC/MS grade (P/N A456-500)
• Fisher Chemical Ammonium Hydroxide, LC/MS grade (P/N A470-250)
• Fisher Chemical Water, Optima™ LC/MS grade (P/N W6500)
• Thermo Scientific™ Pierce™ Formic Acid (P/N PI28905)
• SOLAμ SCX 96-well plate, 2 mg/1 mL (P/N 60209-002) or SOLA SCX cartridges, 10 mg/1 mL (P/N 60109-002)
Protocol
1. Prepare SPE stationary phase for amino acid binding adding by equilibrating with 1 equivalent of methanol (200 µL for SOLAµ 2 mg, or 500 µL for SOLA 10 mg).
2. Wash SPE stationary phase with 1 equivalent of water with 1% formic acid.
3. Acidify sample containing amino acids, for example dilute 500 µL sample with 500 µL 1% formic acid. The sample should be 100% aqueous for optimal binding to stationary phase.
4. Load sample over SPE stationary phase (sample should be applied slowly).
5. Collect flow through from loading step and reapply.
6. Wash SPE stationary phase with 1 equivalent of methanol with 1% formic acid.
7. Elute amino acids from stationary phase with 1 equivalent of 5% ammonium hydroxide in methanol.
8. Analyze your sample.

Related products

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<thead>
<tr>
<th>Description</th>
<th>Part number</th>
</tr>
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<tbody>
<tr>
<td>SOLAµ SCX 96-well plate, 2 mg/1 mL</td>
<td>60209-002</td>
</tr>
<tr>
<td>Positive Pressure Manifold, 96-well plate format</td>
<td>60103-357</td>
</tr>
<tr>
<td>Positive Pressure Manifold, for cartridges, 13 mm tube collection rack</td>
<td>60104-236</td>
</tr>
<tr>
<td>96-well Plate +, glass-coated square 96 well plate</td>
<td>60180-P308</td>
</tr>
<tr>
<td>Thermo Scientific™ WebSeal Mat, 96 well square plate</td>
<td>60180-M122</td>
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</tbody>
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Current versions of product instructions are available at thermofisher.com/chromexpert

Learn more about SOLA and SOLAµ products at thermofisher.com/solaspe

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