STOP wishing you could successfully achieve the results and solve the problems listed above and start actually doing it. The capillary solution (150 x 0.1mm (100µm)) offered within the Onyx portfolio gives those in proteomic and biomarker laboratories capabilities not had before with common silica particle capillary columns.

**Onyx C18 150 x 0.1mm:**
A unique solution for proteomic applications

Onyx capillary HPLC columns are specially engineered by directly forming a C18 bonded monolithic silica gel rod within a 100µm I.D. glass capillary tube. This process creates a column that is highly suited for proteomic applications that require low flow rates, high peak capacities, and low sample-to-sample carryover.

**Applications:**
- Complex proteomic samples with a large number of peptides
- Protein digests with very low-level peptides for LC/MS analysis
- Complex clinical biomarker identification samples
- Differential display analysis using ICAT or other sample comparison technologies
- Two-dimensional LC-LC/MS/MS applications that use reversed phase for the second dimension

**Improved results:**
- More peptide identifications due to the greater peak capacity resulting from high efficiency
- Minimal background noise and longer column lifetimes due to lower carryover
- More method flexibility due to the wider flow rate range made possible by lower backpressures
- Easier coupling to 2-D chromatography methods due to Onyx’s enhanced resistance to salts and other contaminants interfering at the inlet of the column
Tryptic digest of β-Amylase

**Background:** β-Amylase is digested by Trypsin to generate a complex peptide map. No desalting or trap column is used, demonstrating Onyx’s resistance to matrix contaminants.

<table>
<thead>
<tr>
<th>Challenge</th>
<th>Solution</th>
</tr>
</thead>
<tbody>
<tr>
<td>Resolve large numbers of different peptides from a protein digest in a micro capillary format.</td>
<td>The monolithic phase of Onyx delivers high peak efficiency and good peak shape even in a capillary format.</td>
</tr>
<tr>
<td>Maintain separation of peptides when loading in the presence of buffer salts.</td>
<td>The pore structure of Onyx’s monolithic phase allows buffer salts to pass quickly through the column while retaining and separating peptides efficiently across the large surface area of the column.</td>
</tr>
</tbody>
</table>

**Column:** Onyx Monolithic C18  
**Dimensions:** 150 x 0.1mm  
**Order no.:** CH0-7846  
**Mobile phase**  
A: 0.1% TFA in Water  
B: 0.08% TFA#/ 95% Acetonitrile  
**Gradient:** 5-65% B in 60 minutes  
**Flow rate:** 3.0 µL/min  
**Temperature:** Ambient  
**Detection:** UV @ 210nm  
**Sample:** 1. β-Amylase Tryptic Digest
High volume loading of a cytochrome C digest

**Background:** A large volume of a protein digest is directly loaded onto the Onyx column at a high flow rate (10µL/min) without the use of a trapping column. Flow rate is reduced during analysis to improve sensitivity.

**Challenge #1:** Load large volumes of sample quickly without the use of a trap column.

**Solution:** Onyx monolithic silica allows for wide flow rate flexibility resulting in loading at 10µL/min flow rates. A 20µL sample can be directly loaded in less than 2 minutes without the use of a trap column.

**Challenge #2:** Maintain good chromatography at varying flow rates to minimize loading time and maximize analysis time.

**Solution:** The low backpressure and high efficiency of the Onyx monolithic format delivers high efficiency separations across a wide flow rate range (200nL/min-4µL/min). With Onyx, you can use flow rate to optimize the speed and sensitivity of your application.

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**Graphical representation**

<table>
<thead>
<tr>
<th>min</th>
<th>mAU</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>-2</td>
</tr>
<tr>
<td>10</td>
<td>0</td>
</tr>
<tr>
<td>20</td>
<td>2</td>
</tr>
<tr>
<td>30</td>
<td>4</td>
</tr>
<tr>
<td>40</td>
<td>6</td>
</tr>
</tbody>
</table>

**Column:** Onyx Monolithic C18
**Dimensions:** 150 x 0.1mm
**Order no.:** CH0-7646
**Loading:**
**Mobile phase:** 0.1% TFA/5% Acetonitrile/Water
**Flow rate:** 10 µL/min for 2 minutes
**Elution:**
**Mobile Phase:** A: 0.1% TFA in Water  B: 0.08% TFA/ 95% Acetonitrile
**Gradient:** 5 - 85% B in 60 minutes
**Flow rate:** 400 nL/min
**Temperature:** Ambient
**Detection:** UV @ 210nm

**App ID:** 15537
Key Onyx™ capillary features & benefits for proteomic and biomarker applications

1) High peak capacity
   • Onyx columns have high efficiency due to the improved flow characteristics of the monolithic material. Higher efficiency separations deliver greater peak capacity resulting in an increase in peptide identifications.

2) Flow rate flexibility
   • High flow rate flexibility (200nL/min – 10µL/min) allows for rapid loading of sample directly on to the column at high flow rates, while still maintaining good efficiency at both extremes.

3) Low carryover
   • Trap loading is unnecessary with the flow rate flexibility and low carryover of Onyx capillary columns. Eliminating use of a trap columns, removes an additional source of carryover and peak broadening.
   • Compared to typical particle-based columns, Onyx has decreased run-to-run carryover due to the monolithic nature.

4) Low backpressure
   • Low backpressures of the monolithic Onyx capillary column make “peak parking” easier. This is because flow rate changes occur rapidly with less likelihood of losing the peak of interest by pressure drops forcing the peak through.

Onyx™ products for proteomic and biomarker applications

<table>
<thead>
<tr>
<th>Order No.</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>CH0-7641</td>
<td>Onyx Monolithic C18 150 x 0.1 mm</td>
</tr>
</tbody>
</table>

Phenomenex product based on monolithic technology under license from Merck KGaA, Darmstadt, Germany.

Phenomenex products are available worldwide. For the distributor in your country, contact Phenomenex USA, International Department by telephone, fax or e-mail: international@phenomenex.com.

www.phenomenex.com