



# Core-Shell Technology for Proteins and Peptides

*Ultra-High Resolution and Performance on HPLC and UHPLC Systems*



 **phenomenex**<sup>®</sup>  
...breaking with tradition<sup>SM</sup>



# Welcome to the Future of BioSeparations

Introducing **Aeris™**, a specialized line of reversed phase core-shell HPLC / UHPLC columns, built exclusively for the ultra-high performance separation and analysis of proteins and peptides.

These columns can provide improved **resolving power, selectivity, throughput, sensitivity, column lifetime,** and **method flexibility** compared to other fully porous and core-shell columns typically used for bioseparations.

Choose your optimal  
Aeris column

See page 6!

## Aeris WIDEPORE

# p16

Large pore optimized for  
protein diffusion



XB-C18

XB-C8

C4

Multiple selectivities

3.6 μm particle for  
HPLC and UHPLC systems

3.6 μm

## Aeris PEPTIDE

# p26

Small pore optimized  
for peptide diffusion



XB-C18

Ideal surface chemistry  
for resolving peptides

Three scalable particle sizes  
for method and system flexibility

1.7 μm

2.6 μm

3.6 μm



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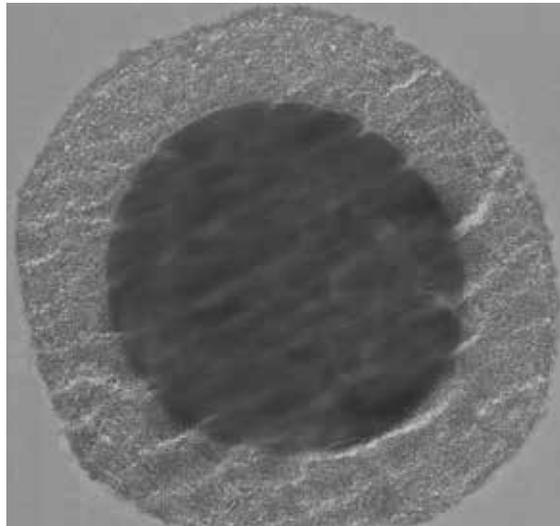
“Aeris had better separation abilities and lower backpressure than other core-shell reversed phase columns I've tried with proteins. I had very good technical and customer support throughout the entire process! I'm very glad to have switched to Phenomenex columns!”  
-LYNN PRUISNER, TECHNOLOGY COMPANY

# Core-Shell Particles Precision Engineered for Protein and Peptide Separations

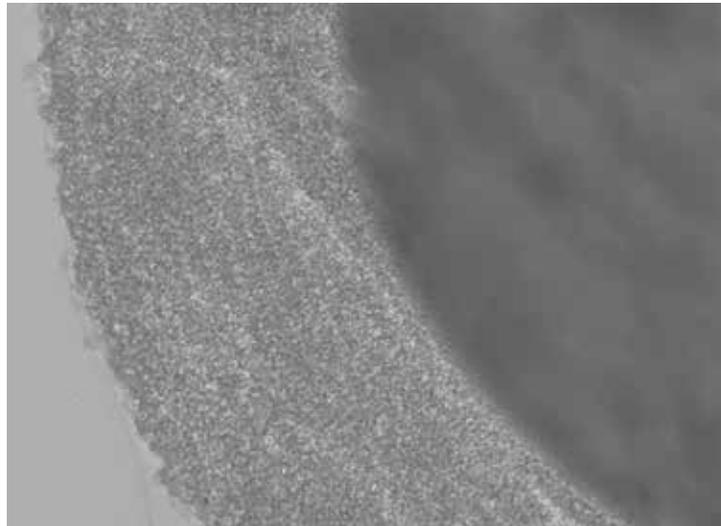
Core-shell particle technology provides **striking increases in peak capacity and resolution** at lower backpressures, giving chromatographers the ability to achieve ultra-high performance on ANY system, HPLC or UHPLC.

A uniform porous silica layer is grown around a solid, spherical silica core, providing effective retention and selectivity with improved resolution, speed, and recovery. Next, optimizing the pore size and shell thickness for intact proteins or smaller peptide fragments provides well-defined depth penetration of biomolecules leading to **maximum separation power**.

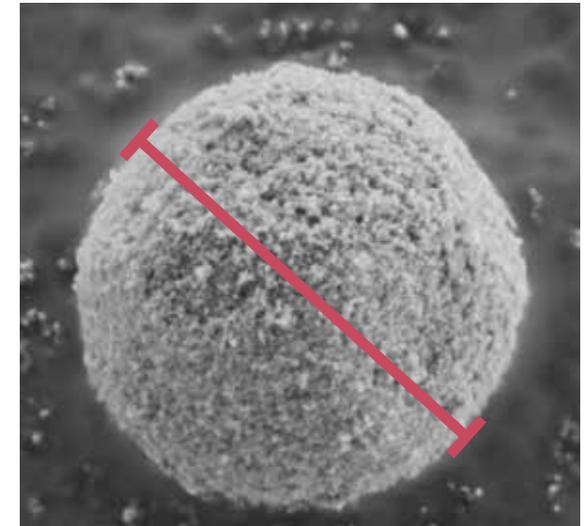
## TEM and SEM of Aeris™ PEPTIDE 3.6µm Core-Shell Particles



**Cross section of an  
Aeris core-shell particle**



**Magnified cross section of the porous "shell"**



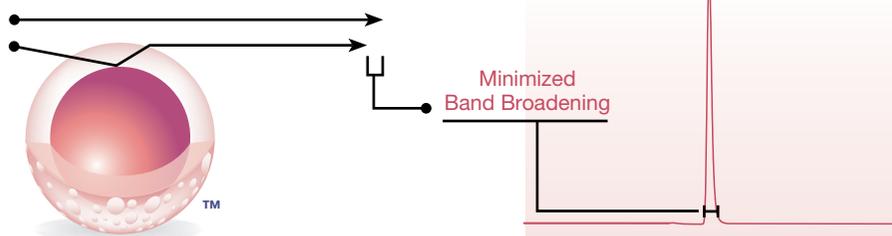
**Uniform particle size and shape**

The precise architecture of core-shell particles provides dramatic leaps in performance in two important ways:

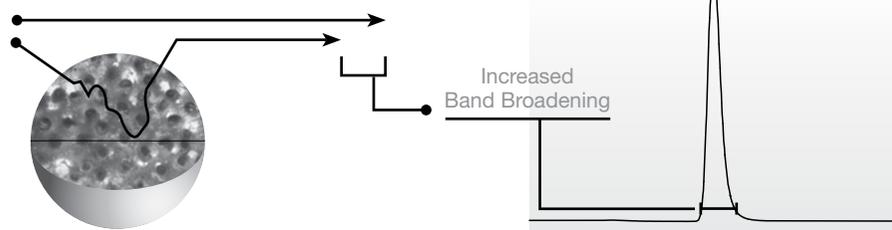
1

The thin, porous layer, or “shell”, decreases the diffusion path length, thus reducing the time it takes for biomolecules to adsorb/desorb into and out of the particle.

Aeris Core-Shell Particle



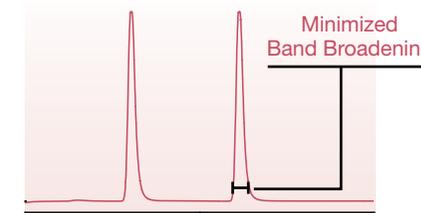
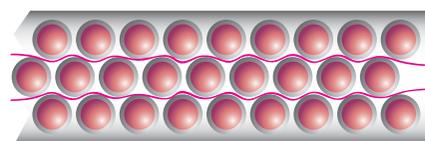
Fully Porous Particle



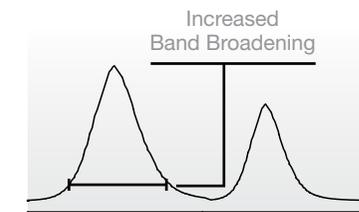
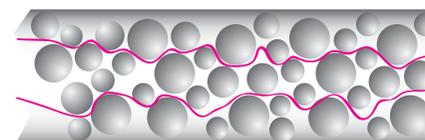
2

Uniform sizing and shape of the particles along with tight packing specifications reduces losses in efficiency and performance due to band broadening.

Aeris Core-Shell Particles



Fully Porous Particles



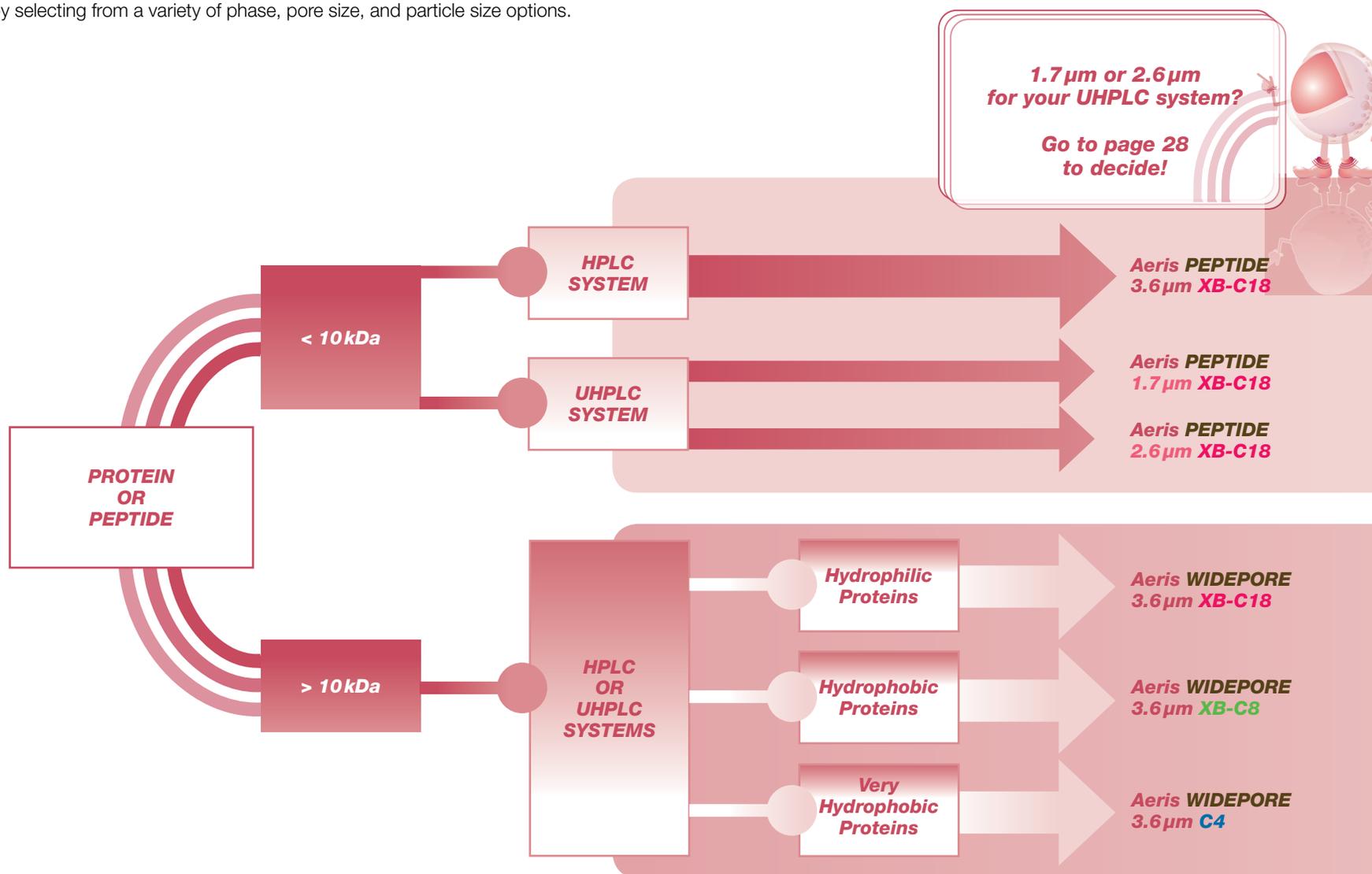
The result is

- **3.6 µm core-shell particles** that can perform like sub-2 µm columns on both HPLC and UHPLC systems at a fraction of the pressure
- **1.7 µm and 2.6 µm core-shell particles** that can provide higher peak capacities compared to fully porous sub-2 µm columns on UHPLC systems

# Selecting the Optimal Aeris Column for Your Applications



Aeris™ core-shell columns are designed for the separation of complex protein and peptide mixtures. Chromatographers can easily narrow down the column(s) that has a high probability of success for their separation by selecting from a variety of phase, pore size, and particle size options.



# Optimizing an Aeris Column With Your Method and System

## Aeris WIDEPORE

Because of the reduced hydrophobicity compared to fully porous 300Å columns, one should start gradients with reduced organic concentrations to increase retention and improve peak shape of polar proteins and peptides. Shallower gradients compared to fully porous columns may also be appropriate.

## Aeris PEPTIDE

For increased resolving power, use a longer column (preferably a 250 mm) to maximize separation. For specific peptide map applications, one can also use different gradient slope segments to “stretch” or “compress” regions of a peptide map.

## Flow Rate and Column Length

Start method development using column dimensions and flow rates similar to existing protein or peptide separation methods. For higher throughput, use shorter column lengths and higher flow rates for minimal impact on resolution. For maximum resolution, use longer columns and shallower gradients.

| Column ID         | Flow Rate      |
|-------------------|----------------|
| 4.6 mm ID columns | 0.8-2.0 mL/min |
| 2.1 mm ID columns | 0.2-0.5 mL/min |

## HPLC System Optimization

Aeris 2.6 μm and 3.6 μm core-shell columns operate comfortably within the pressure limits of conventional HPLC systems and meet or exceed the performance of sub-2 μm fully porous particle columns on UHPLC-systems. To maximize the benefits of your Aeris Core-Shell columns, one should investigate:

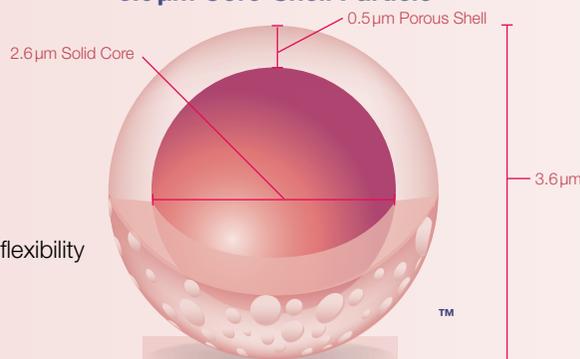
- Minimizing peak dispersion by reducing the system dwell volume between the injector and detector
- Optimizing detector settings by adjusting the scan rate and/or time constant to the fastest practical setting such that signal-to-noise ratio (S/N) is not adversely affected

## Aeris PEPTIDE

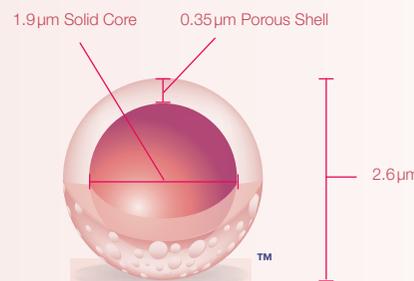
Recommended for the separation of low molecular weight peptides and peptide mapping.

- XB-C18 chemistry best suited for resolving peptides
- 1.7 μm, 2.6 μm and 3.6 μm particles for method development flexibility
- Small pore optimized for peptide diffusion

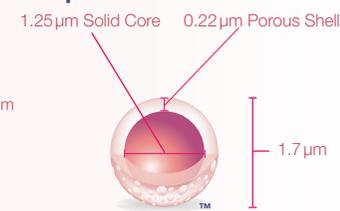
### 3.6 μm Core-Shell Particle



### 2.6 μm Core-Shell Particle



### 1.7 μm Core-Shell Particle

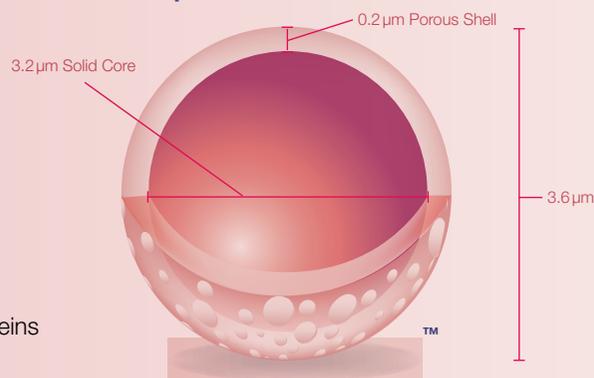


## Aeris WIDEPORE

Recommended for the separation of intact proteins and large oligonucleotides.

- XB-C18, XB-C8, and C4 phases for alternate selectivities
- 3.6 μm particle for system flexibility
- Thin shell optimized for fast protein adsorption/desorption
- High pore permeability for improved separation of very large proteins (up to 400 kDa)

### 3.6 μm Core-Shell Particle



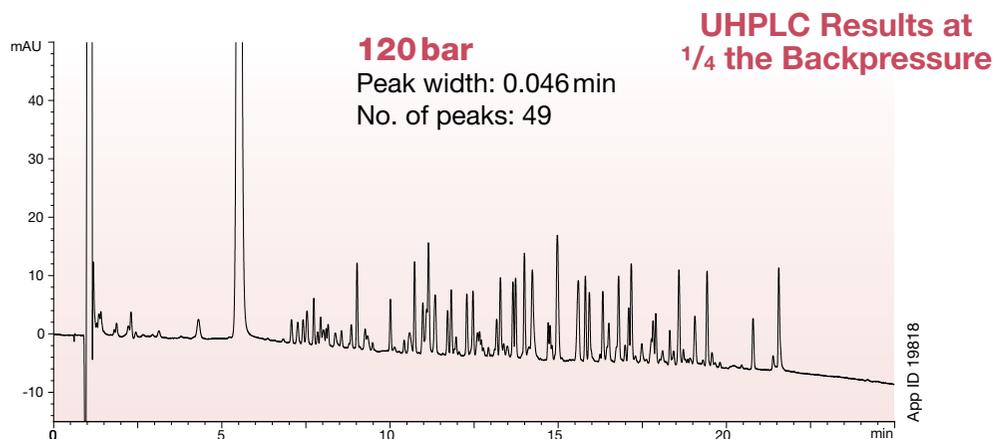
**Aeris WIDEPORE  
XB-C18 and  
Aeris PEPTIDE XB-C18  
make a perfect pair  
for peptide mapping.  
See p. 32 for more details.**

# Improve Resolution on ANY System by Leveraging Low Backpressure

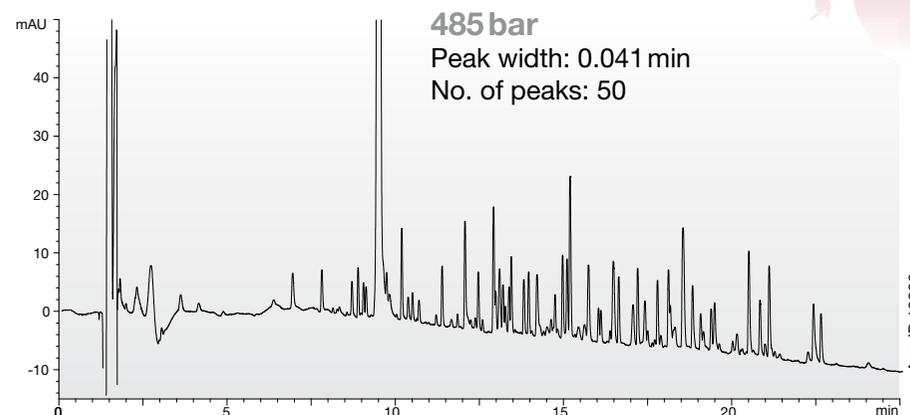
Aeris™ PEPTIDE and Aeris WIDEPORE 3.6µm columns can **perform like sub-2µm columns at a fraction of the backpressure**. This allows chromatographers to utilize the resolving power of longer length (or coupled) columns without exceeding the pressure limits of their HPLC system. Scientists analyzing proteins and peptides can now have ultra-high resolution on HPLC or UHPLC systems.

## Sub-2µm Performance at a Fraction of the Backpressure

### Aeris WIDEPORE 3.6µm XB-C18



### Waters® ACQUITY® BEH300 1.7µm C18



#### Conditions for both columns:

**Column:** Aeris WIDEPORE 3.6µm XB-C18  
ACQUITY® BEH300 1.7µm C18  
**Dimensions:** 150 x 2.1 mm  
**Mobile Phase:** A: Water with 0.1% TFA  
B: Acetonitrile with 0.1% TFA  
**Gradient:** A/B (65:35) for 3 min to A/B (35:65) over 30 min

**Flow Rate:** 0.3 mL/min  
**Temperature:** 40 °C  
**Injection Volume:** 20 µL  
**Instrument:** Agilent® 1200SL  
**Detection:** UV @ 214 nm (ambient)  
**Sample:** BSA (Bovine Serum Albumin) Tryptic Digest

**Using a UHPLC system?**

**Try Aeris PEPTIDE 1.7µm columns for ultra-high efficiency peptide maps and stability up to 1,000 bar.**

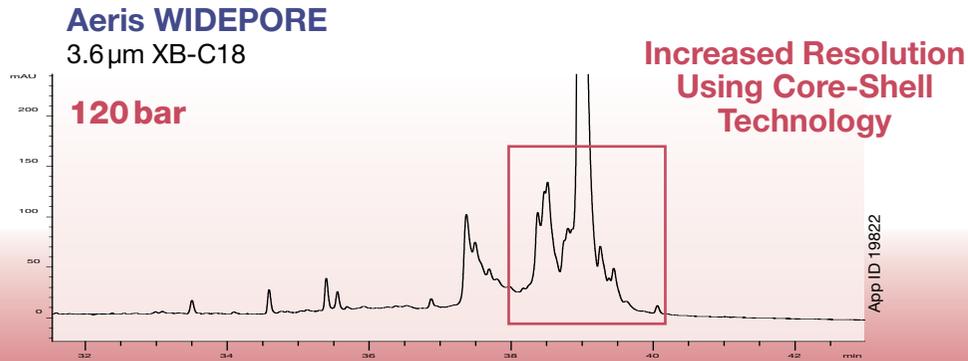
**See page 11!**



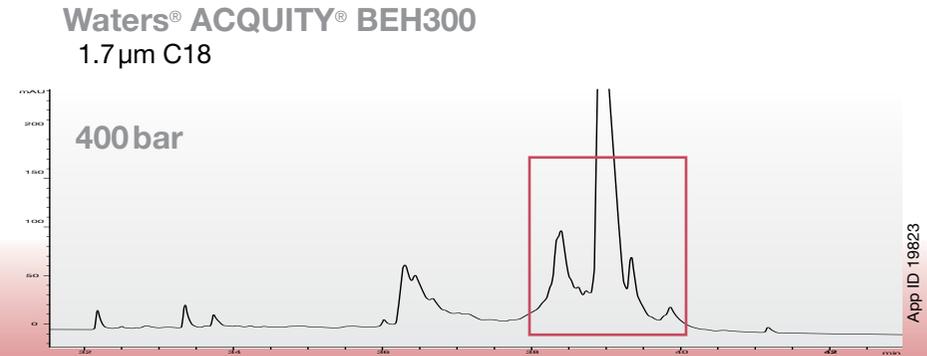
ACQUITY and Waters are registered trademarks, and BEH Technology is a trademark of Waters Corporation. Phenomenex is not affiliated with Waters Corporation. Study was performed using new columns and, to the extent possible, identical experimental conditions were applied. Comparative separations may not be representative of all applications.



**Utilize Long Columns to Maximize Resolution on UHPLC Systems**



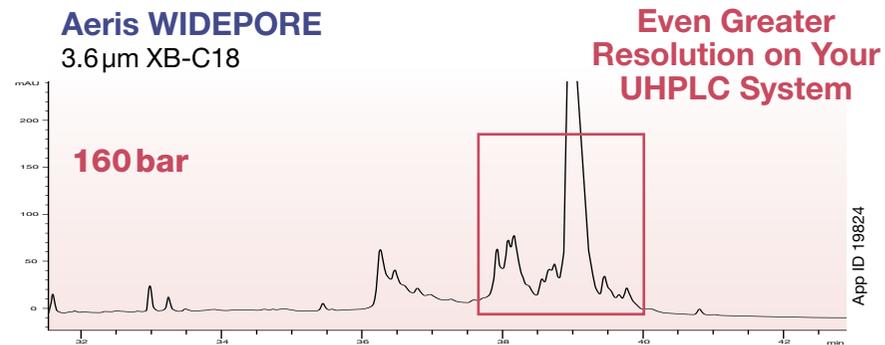
**150 x 2.1 mm**



**150 x 2.1 mm**



**250 x 2.1 mm**



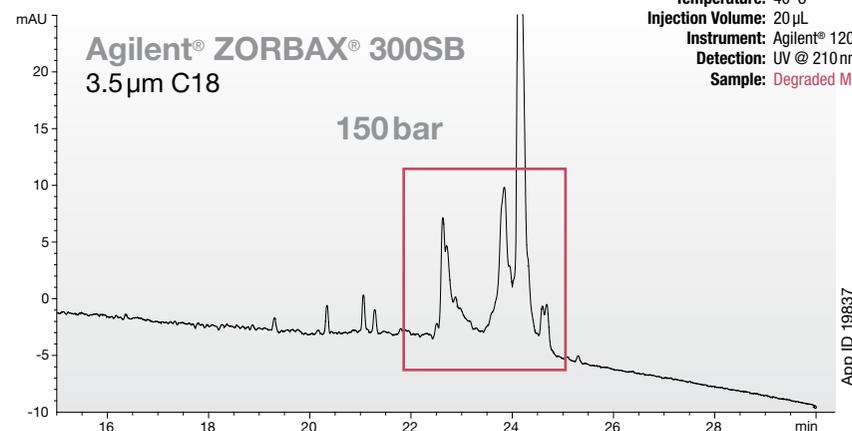
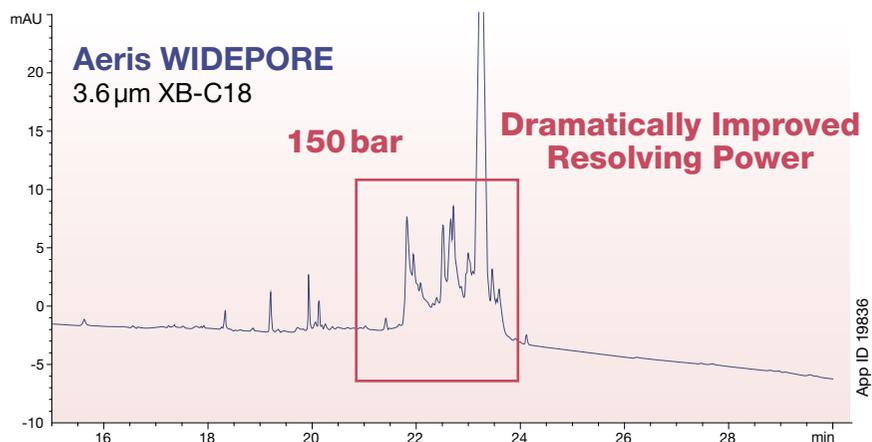
- Conditions for all columns:**
- Column:** Aeris WIDEPORÉ 3.6µm XB-C18  
ACQUITY® BEH300 1.7µm C18
  - Dimensions:** as noted in chromatogram
  - Mobile Phase:** A: Water with 0.1% TFA  
B: Acetonitrile
  - Gradient:** A/B (90:10) for 5 min to A/B (50:50) over 45 min
  - Flow Rate:** 0.2 mL/min
  - Temperature:** 22°C
  - Injection Volume:** 20 µL
  - Instrument:** Agilent® 1200SL
  - Detection:** UV @ 210 nm
  - Sample:** Degraded Myoglobin

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# Achieve UHPLC Performance on HPLC Systems by Replacing 3 $\mu\text{m}$ and 5 $\mu\text{m}$ Columns

The innovative structure of 3.6  $\mu\text{m}$  Aeris™ core-shell particles was specially designed to provide sub-2  $\mu\text{m}$  performance at backpressures similar to fully porous 3  $\mu\text{m}$  and 5  $\mu\text{m}$  particles. Aeris columns can deliver increased resolution for existing protein and peptide separations performed on fully porous 3  $\mu\text{m}$  and 5  $\mu\text{m}$  columns, using the same HPLC system!

## Upgrade Existing Methods on 3 $\mu\text{m}$ and 5 $\mu\text{m}$ Fully Porous Columns to Aeris Core-Shell Technology



**Conditions for both columns:**  
**Column:** Aeris WIDEPORE 3.6  $\mu\text{m}$  XB-C18  
ZORBAX® 300SB 3.5  $\mu\text{m}$  C18  
**Dimensions:** 150 x 4.6 mm  
**Mobile Phase:** A: Water with 0.1 % TFA  
B: Acetonitrile with 0.1 % TFA  
**Gradient:** A/B (97:3) for 3 min to A/B (35:65) over 30 min  
**Flow Rate:** 1.5 mL/min  
**Temperature:** 40 °C  
**Injection Volume:** 20  $\mu\text{L}$   
**Instrument:** Agilent® 1200SL  
**Detection:** UV @ 210 nm (ambient)  
**Sample:** Degraded Myoglobin

Agilent and ZORBAX are registered trademarks of Agilent Technologies, Inc. Phenomenex is not affiliated with Agilent Technologies, Inc. Study was performed using new columns and, to the extent possible, identical experimental conditions were applied. Comparative separations may not be representative of all applications.

**Improving your  
current method is fast and  
easy with an  
Aeris core-shell column.**



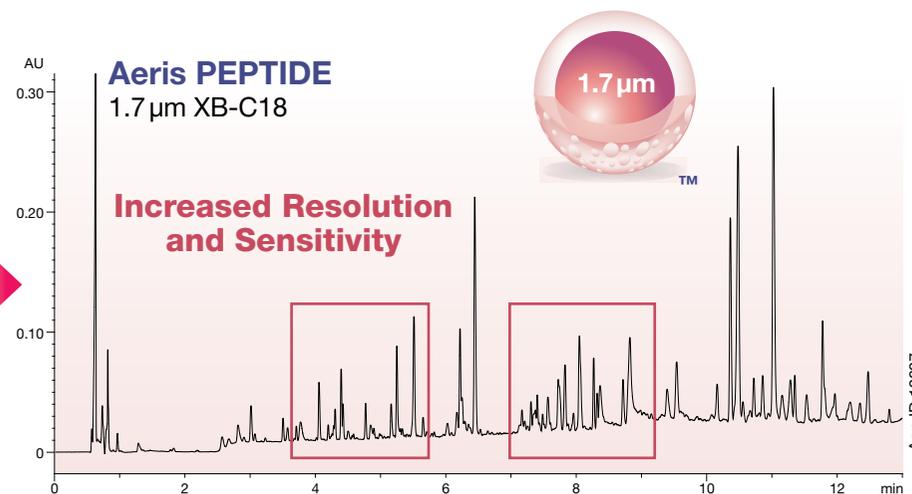
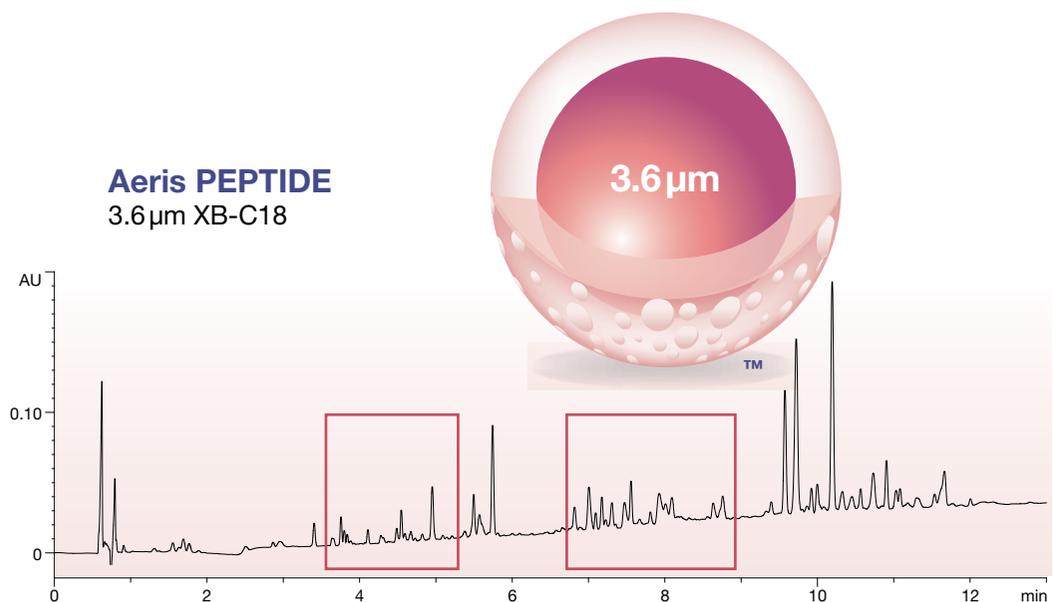


# Increase Efficiency on UHPLC Systems with Sub-2 $\mu\text{m}$ Core-Shell Particles

For labs that have adopted higher pressure capable UHPLC instruments, Aeris PEPTIDE 1.7  $\mu\text{m}$  and 2.6  $\mu\text{m}$  core-shell columns are an excellent solution for ultra-high resolution peptide and peptide mapping separations. Core-shell particle technology combined with a sub-2  $\mu\text{m}$  particle size results in extremely high efficiencies that scientists can use to pull apart critical peaks.

## Ultra-High Resolution Achieved with 1.7 $\mu\text{m}$ Core-Shell Technology

**Conditions for both columns:**  
**Column:** Aeris PEPTIDE 3.6  $\mu\text{m}$  XB-C18  
 Aeris PEPTIDE 1.7  $\mu\text{m}$  XB-C18  
**Dimensions:** 150 x 2.1 mm  
**Part Nos.:** 00F-4507-AN  
 00F-4506-AN  
**Mobile Phase:** A: Water with 0.1 % TFA  
 B: Acetonitrile with 0.08 % TFA  
**Gradient:** A/B (97:3) for 1.5 min to A/B (60:40) over 11 min to  
 A/B (5/95) for 1 min  
**Flow Rate:** 0.5 mL/min  
**Temperature:** 40 °C  
**Injection Volume:** 5  $\mu\text{L}$   
**Instrument:** Agilent® 1200SL  
**Detection:** UV @ 214 nm (ambient)  
**Sample:** Alpha-Casein Tryptic Digest

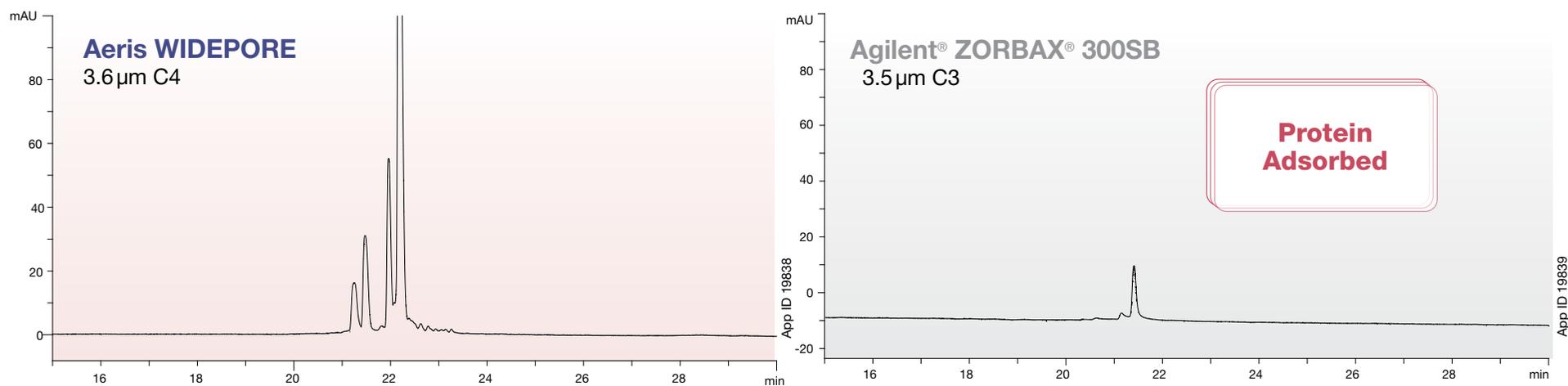


Study was performed using new columns and, to the extent possible, identical experimental conditions were applied. Comparative separations may not be representative of all applications.

# Minimize Adsorption and Maximize Recoveries for Accurate Results

Aeris™ phase chemistries and bonding technology create a highly inert surface, leading to greatly reduced irreversible adsorption, higher recoveries, and sharper, narrower peaks, providing high quality and accurate results for each consecutive analysis.

## Maximize Recoveries of Hydrophobic Proteins



### Conditions for both columns:

**Column:** Aeris WIDEPORE 3.6 μm C4  
ZORBAX® 300SB 3.5 μm C3

**Dimensions:** 150 x 2.1 mm

**Mobile Phase:** A: Water with 0.1 % TFA  
B: Acetonitrile with 0.1 % TFA

**Gradient:** A/B (97:3) to A/B (35:65) over 45 min

**Flow Rate:** 0.3 mL/min

**Temperature:** 40 °C

**Injection Volume:** 20 μL

**Instrument:** Agilent® 1200

**Detection:** UV @ 214 nm (ambient)

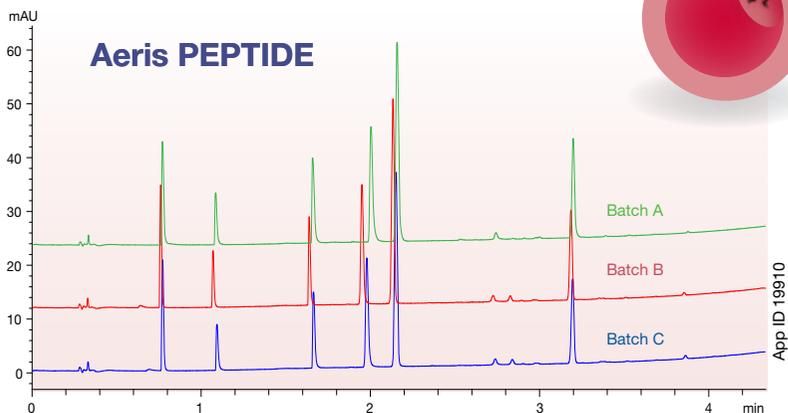
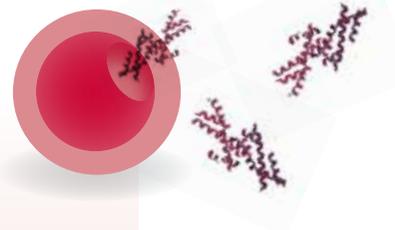
**Sample:** Human Epidermal Growth Factor

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# Tightly Controlled Quality for Reproducible Data

Every Aeris column and batch of media undergoes quality assurance tests for particle size distribution (both solid core and final particle), surface coverage, carbon load, pore diameter, pore size distribution, and other parameters to ensure **exceptional reproducibility for worry-free methods and confident results.**

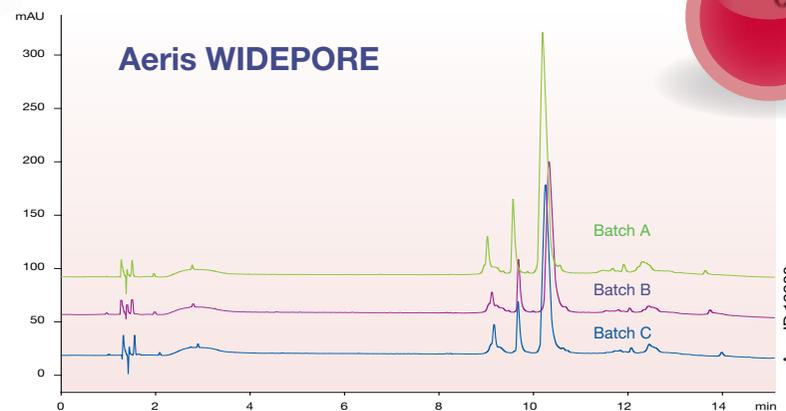
## Batch-to-Batch Reproducibility



App ID 19910

**Column:** Aeris PEPTIDE 1.7  $\mu$ m XB-C18  
**Dimensions:** 50 x 4.6 mm  
**Part No.:** 00B-4506-E0  
**Mobile Phase:** A: Water with 0.1% Formic Acid  
 B: Acetonitrile with 0.1% Formic Acid  
**Gradient:** A/B (95:5) to A/B (5:95) over 4 min  
**Flow Rate:** 1.85 mL/min  
**Temperature:** 30 °C  
**Injection Volume:** 0.4  $\mu$ L  
**Detection:** UV @ 254 nm (ambient)  
**Sample:** Selectivity Test Mixture

## Batch-to-Batch Reproducibility



App ID 19908

**Column:** Aeris WIDEPOR 3.6  $\mu$ m XB-C18  
**Dimensions:** 150 x 4.6 mm  
**Part No.:** 00F-4482-E0  
**Mobile Phase:** A: Water with 0.1% Formic Acid  
 B: Acetonitrile with 0.085% Formic Acid  
**Gradient:** A/B (95:5) to A/B (5:95) over 20 min  
**Flow Rate:** 1.0 mL/min  
**Temperature:** 40 °C  
**Injection Volume:** 0.2  $\mu$ L  
**Detection:** UV @ 210 nm (ambient)  
**Sample:** Mouse IgG

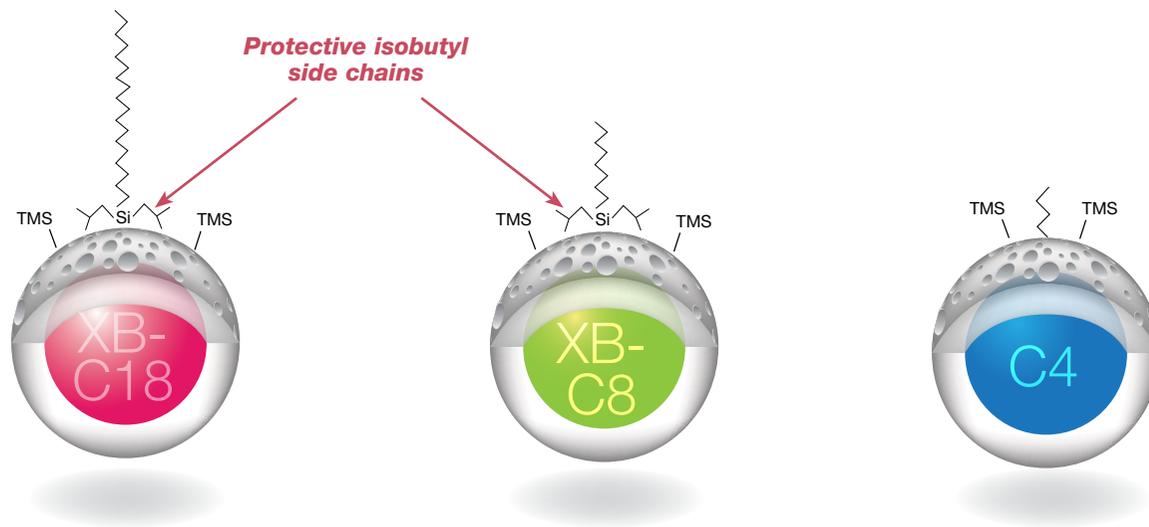
# Greater Method Flexibility with Specialty Surface Chemistries

Aeris™ WIDEPORE columns are available in three surface chemistries (XB-C18, XB-C8, C4) to satisfy applications of all types, ranging from sticky, intact proteins to complex protein digests.

Aeris PEPTIDE columns utilize the XB-C18 chemistry, as it is optimal for peptides and peptide mapping applications.

The unique, sterically protected XB surface ligands are designed by bonding bulky isobutyl chains beside the alkyl chains, and then fully end-capping the surface to cover any remaining exposed silanols.

An added benefit of XB chemistry is its high temperature stability, which allows one to use elevated column temperatures up to 90 °C for improved peak shape and recovery.



**The Aeris WIDEPORE C4 phase does not use the XB chemistry, as shorter chain alkyl phases have higher bonding densities, thus providing steric hindrance. This means that chemical stability, inertness, and low bleed are maintained. The Aeris WIDEPORE C4 phase is an excellent complement to the other phases, and is also temperature stable to 90 °C**



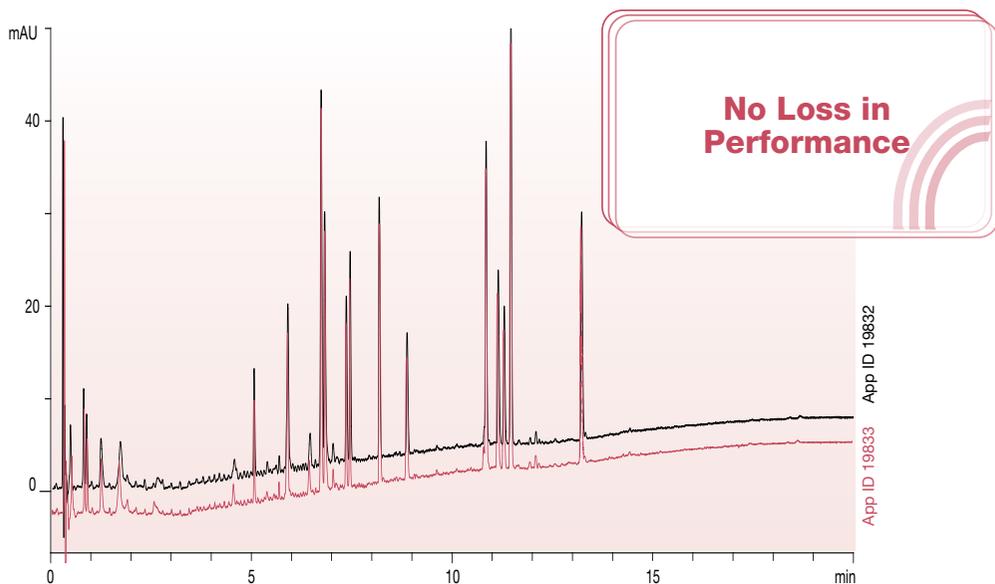
# Long Column Lifetimes Under Extreme Method Conditions

# Low Column Bleed for Amplified Mass Spec (MS) Sensitivity

Aeris columns provide temperature stability up to 90 °C, and pH stability from 1.5 - 9, giving ample flexibility for method development and excellent column lifetime.

Aeris columns show no significant phase bleed under LC/MS conditions, making them very suitable for protein and peptide analysis. Chemists can be assured accurate, dependable, and consistent results, time and time again.

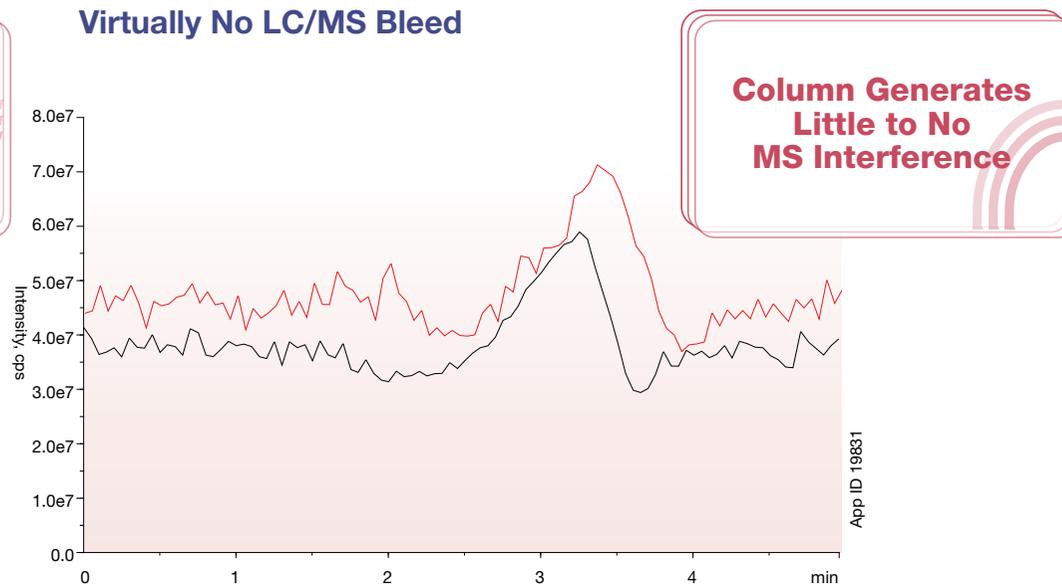
## Over 1,000 Injections at 90 °C



**Column:** Aeris WIDEPORE 3.6 µm XB-C18  
**Dimensions:** 50 x 4.6 mm  
**Part No.:** 00B-4282-E0  
**Mobile Phase:** A: Water with 0.1 % TFA  
 B: Acetonitrile with 0.1 % TFA  
**Gradient:** A/B (97:3) for 3 min, then to A/B (35:65) over 20 min

**Flow Rate:** 1.5 mL/min  
**Temperature:** 90 °C  
**Injection Volume:** 10 µL  
**Detection:** UV @ 214 nm (ambient)  
**Sample:** Apomyoglobin Digest

## Virtually No LC/MS Bleed



**Column:** Aeris WIDEPORE 3.6 µm XB-C18  
**Dimensions:** 50 x 2.1 mm  
**Part No.:** 00B-4282-AN  
**Mobile Phase:** A: Water with 0.1 % Formic Acid  
 B: Acetonitrile with 0.1 % Formic Acid  
**Gradient:** A/B (95:5) for 2.5 min, to A/B (5:95) hold for 0.5 min, then re-equilibrate

**Flow Rate:** 0.5 mL/min  
**Temperature:** 25 °C  
**Detection:** MS (API 4000™)  
 Positive Ion Mode  
 Q1 scan from 75 to 800 amu  
**Sample:** Blank

Study was performed using new columns and, to the extent possible, identical experimental conditions were applied. Comparative separations may not be representative of all applications.

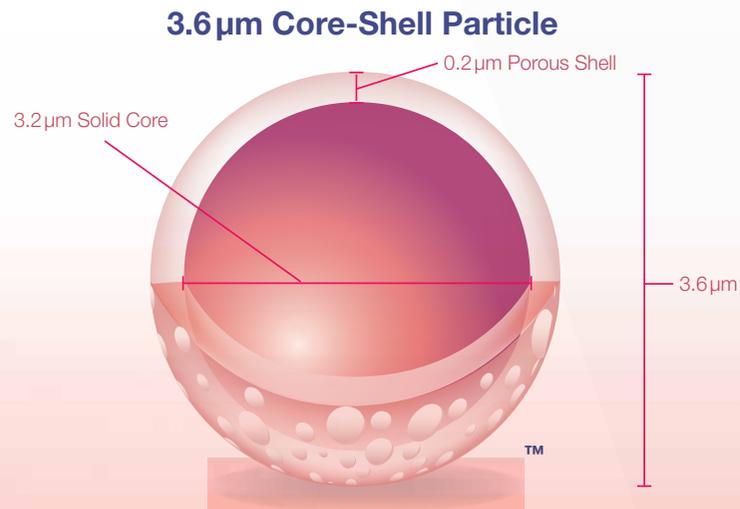
# Aeris™ WIDEPORE Columns

## for Intact Protein and Large Polypeptide Separations

Aeris WIDEPORE columns are packed with 3.6µm core-shell particles that are specially engineered with a thin porous shell, large pores, and sterically protected XB surface chemistry to address the inherent separation challenges of proteins and large peptides. This unique mix of features results in low backpressures, fast rates of diffusion, and excellent selectivity, generating exceptional chromatographic resolution on both HPLC and UHPLC systems.

### Recommended for...

- Protein structural characterization
- Stability indicating assays
- Post-translational modification identification
- PEGylated proteins, antibodies, biosimilars, etc.
- Impurity profiling
- Alternate peptide map selectivity
- Large oligonucleotides





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# Aeris WIDEPORE

**p. 18 Easy Method Development with Three Selectivities**

**p. 20 Maximize HPLC and UHPLC Resolving Power**

**p. 22 Applications**

“The Aeris WIDEPORE column has given our company the opportunity to separate 2 forms of a protein (PEGylated & non-PEGylated). Prior to using Aeris the 2 peaks demonstrated little or no resolution. However by using the Aeris column the 2 peaks are separated by 5 minutes which is excellent.”

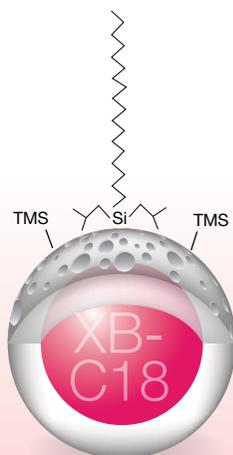
-LARGE PHARMACEUTICAL COMPANY

“Started using the Aeris WIDEPORE XB-C18 and XB-C8 for oligonucleotides and aptamers with excellent results! Very good peak shapes and excellent plate counts on these columns. Really nice to see all of the peaks present in the samples w/o a very long run time. Columns seem to be very stable and have very reasonable backpressures!”

-HEALTHCARE PRODUCTS COMPANY

# Easy Method Development with Three Selectivities

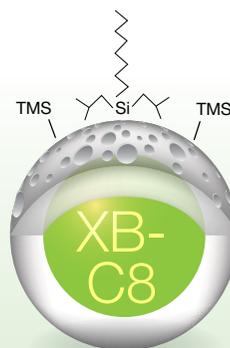
Aeris™ WIDEPORÉ 3.6µm Core-Shell Stationary Phases:



**XB-C18**

**Maximum hydrophobicity  
recommended for:**

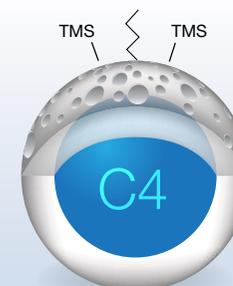
- Proteins
- Hydrophilic proteins
- PEGylated proteins
- High temperature separations
- Alternative selectivity for peptide mapping



**XB-C8**

**Moderate hydrophobicity  
recommended for:**

- Proteins
- Moderately hydrophobic proteins
- Monoclonal antibodies
- Glycosylated proteins
- High temperature separations



**C4**

**Low hydrophobicity  
recommended for:**

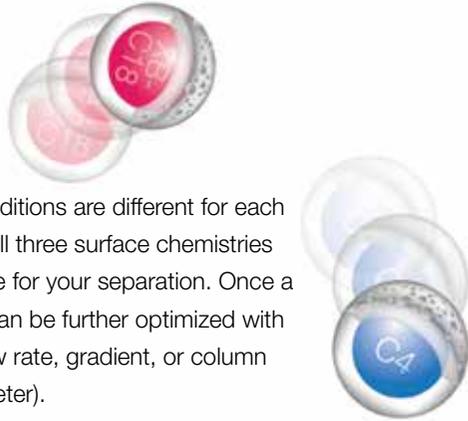
- Very large proteins
- Very hydrophobic proteins
- Membrane proteins
- Least retentive

Want more information on  
the novel XB chemistry?

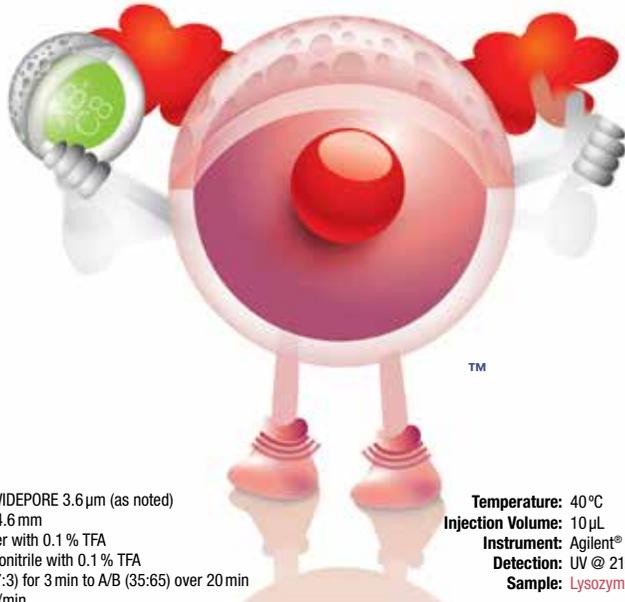
**See page 14!**



# Easy Method Development with Three Selectivities



Because optimal separation conditions are different for each protein, we suggest evaluating all three surface chemistries to uncover the most suitable one for your separation. Once a phase is selected, the method can be further optimized with tweaks to the mobile phase, flow rate, gradient, or column dimension (length, internal diameter).

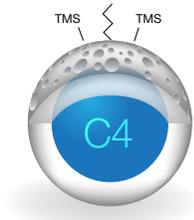
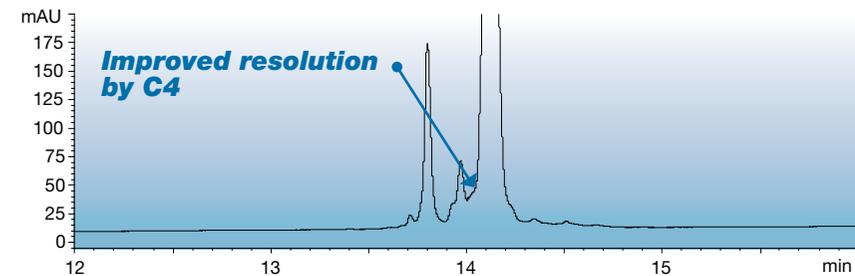
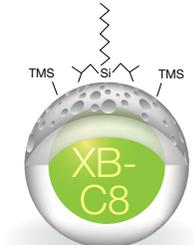
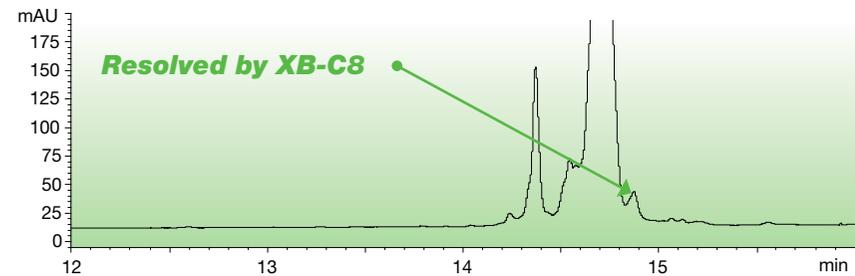
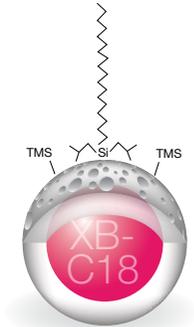
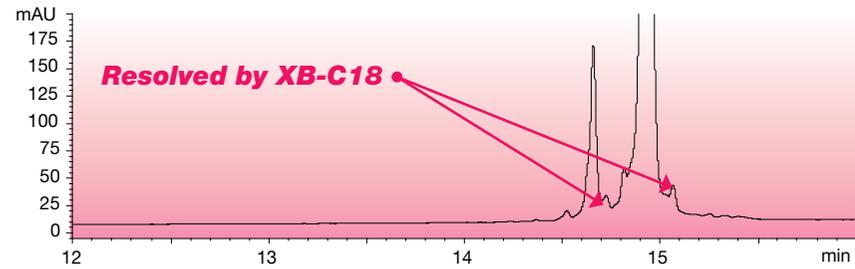


**Conditions for all columns:**

**Column:** Aeris WIDEPORE 3.6 μm (as noted)  
**Dimensions:** 100 x 4.6 mm  
**Mobile Phase:** A: Water with 0.1 % TFA  
 B: Acetonitrile with 0.1 % TFA  
**Gradient:** A/B (97:3) for 3 min to A/B (35:65) over 20 min  
**Flow Rate:** 1.5 mL/min

**Temperature:** 40 °C  
**Injection Volume:** 10 μL  
**Instrument:** Agilent® 1200  
**Detection:** UV @ 214 nm (ambient)  
**Sample:** Lysozyme (1 mg/mL)

## Aeris Phase Selectivity Differences



Study was performed using new columns and, to the extent possible, identical experimental conditions were applied. Comparative separations may not be representative of all applications.

# Maximize HPLC and UHPLC Resolving Power with Unique 3.6 $\mu\text{m}$ Core-Shell Particle

3.6  $\mu\text{m}$  core-shell technology combined with inert surface chemistries and tight packing specifications results in Aeris™ WIDEPORE columns **delivering exceptional resolving power at significantly lower backpressures**. Chromatographers now have the ability to generate higher quality data than typically produced by columns packed with fully porous particles for every protein analysis – on HPLC or UHPLC systems.

#### Conditions for both columns:

**Column:** ACQUITY® BEH300 1.7  $\mu\text{m}$  C4  
Aeris WIDEPORE 3.6  $\mu\text{m}$  C4

**Dimensions:** 150 x 2.1 mm

**Mobile Phase:** A: Water with 0.1% TFA  
B: Acetonitrile with 0.1% TFA

**Gradient:** A/B (97:3) to A/B (35:65) over 45 min

**Flow Rate:** 0.3 mL/min

**Temperature:** 40 °C

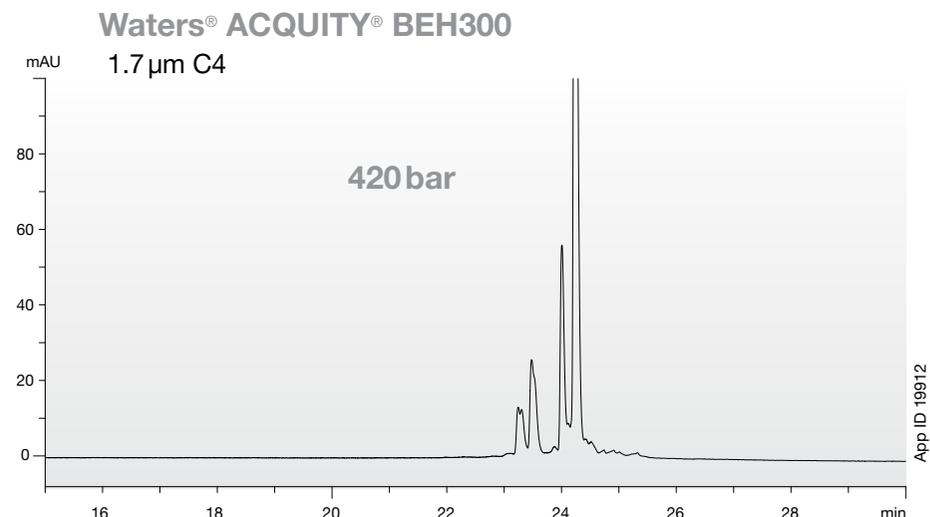
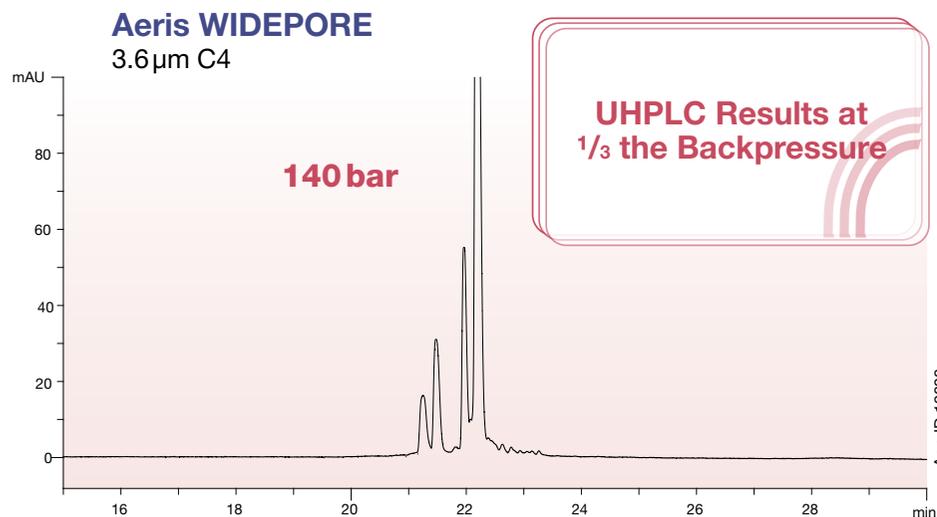
**Injection Volume:** 10  $\mu\text{L}$

**Instrument:** Agilent® 1200

**Detection:** UV @ 214 nm (ambient)

**Sample:** Human Epidermal Growth Factor (EGF)

## Performance Equivalent to sub-2 $\mu\text{m}$ Particle at Low Backpressure



Waters and ACQUITY are registered trademarks, and BEH Technology is a trademark of Waters Corporation. Phenomenex is not affiliated with Waters Corporation. Study was performed using new columns and, to the extent possible, identical experimental conditions were applied. Comparative separations may not be representative of all applications.

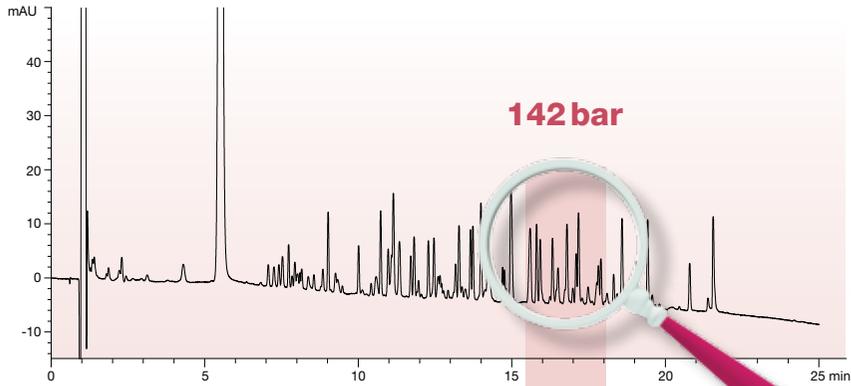


Increase Column Length to Improve Resolving Power

150 x 2.1 mm



Aeris WIDEPORE  
3.6 μm XB-C18

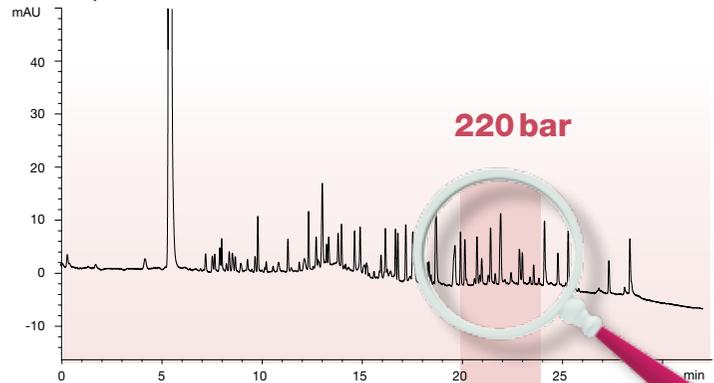


150 mm Length  
Zoom-In

250 x 2.1 mm



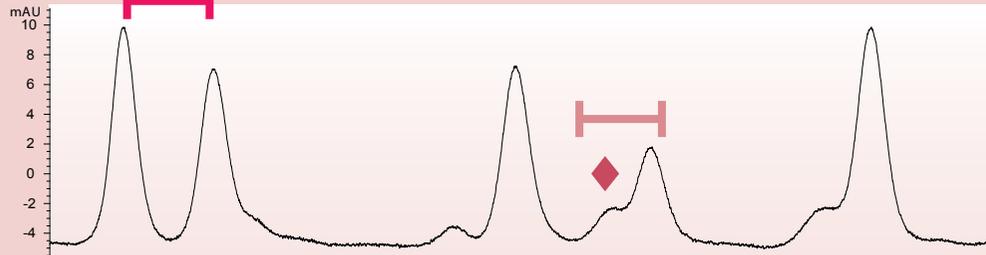
Aeris WIDEPORE  
3.6 μm XB-C18



250 mm Length  
Zoom-In

Conditions for both columns:

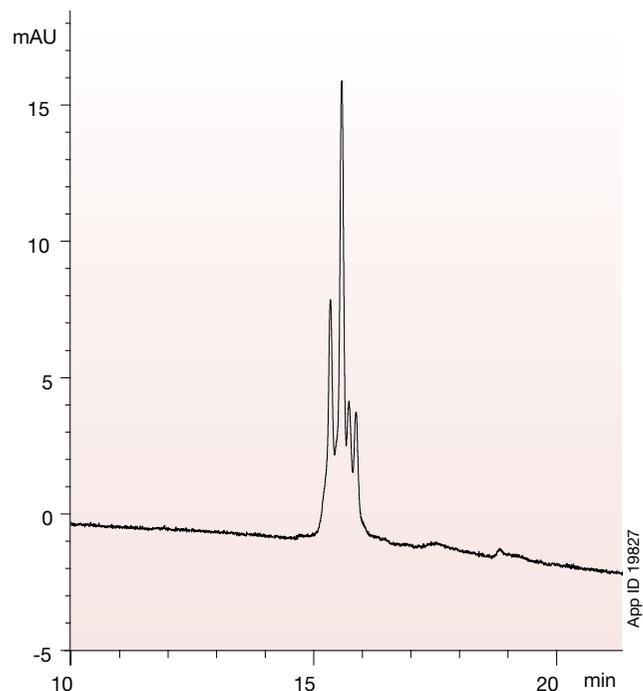
- Column: Aeris WIDEPORE 3.6 μm XB-C18
- Dimensions: as noted
- Mobile Phase: A: Water with 0.1 % TFA  
B: Acetonitrile with 0.1 % TFA
- Gradient: A/B (97:3) for 3 min to A/B (35:65) over 30 min
- Flow Rate: 0.3 mL/min
- Temperature: 40 °C
- Injection Volume: 25 μL
- Instrument: Agilent® 1200
- Detection: UV @ 214 nm (ambient)
- Sample: BSA (Bovine Serum Albumin) Digest



# Applications

## Intact Protein Characterization

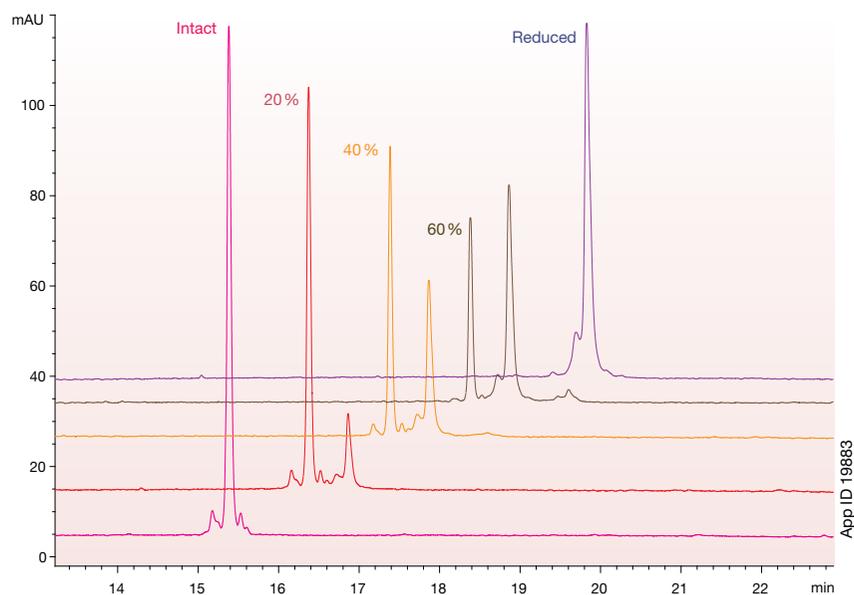
### Biosimilar Impurity Quantitation



**Column:** Aeris™ WIDEPORE 3.6µm XB-C8  
**Dimensions:** 150 x 4.6 mm  
**Part No.:** OOF-4481-E0  
**Mobile Phase:** A: Water with 0.1 % TFA  
B: Acetonitrile with 0.1 % TFA  
**Gradient:** A/B (70:30) to A/B (35:65) over 30 min

**Flow Rate:** 1.0 mL/min  
**Temperature:** 22°C  
**Injection Volume:** 5 µL  
**Instrument:** Agilent® 1200  
**Detection:** UV @ 214 nm (ambient)  
**Sample:** Interferon alpha-2a

### Protein Reduction



**Column:** Aeris WIDEPORE 3.6µm C4  
**Dimensions:** 150 x 4.6 mm  
**Part No.:** OOF-4486-E0  
**Mobile Phase:** A: Water with 0.1 % TFA  
B: Acetonitrile with 0.1 % TFA  
**Gradient:** A/B (97:3) for 3 min to A/B (35:65) over 30 min  
**Flow Rate:** 1.2 mL/min  
**Temperature:** 22°C  
**Injection Volume:** 20 µL  
**Instrument:** Agilent 1200 SL  
**Detection:** UV @ 214 nm (ambient)  
**Sample:** RNase subject to reduction  
100 % intact  
20 % reduced  
40 % reduced  
60 % reduced  
100 % reduced

**Aeris WIDEPORE 3.6µm C4**  
successfully monitors peak  
shifts due to differences in  
protein shape

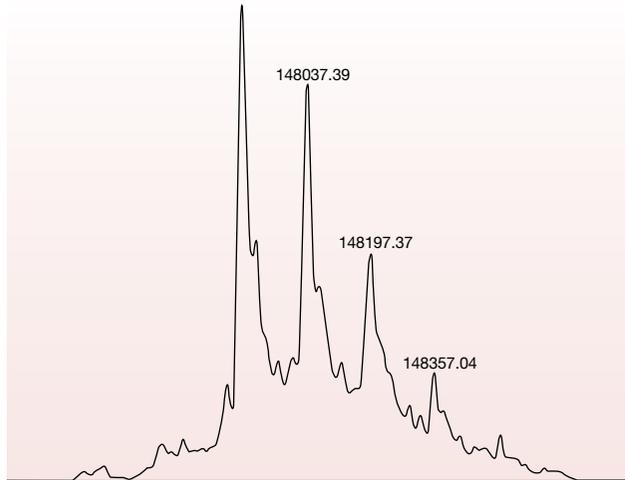




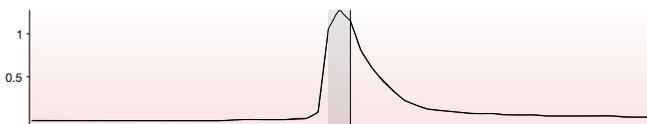
# Applications

## Intact Monoclonal Antibody (mAb) Separation

### Human mAb



App ID 19846



App ID 19846

**Column:** Aeris WIDEPORE 3.6 μm XB-C18  
**Dimensions:** 50 x 2.1 mm  
**Part No.:** 00B-4482-AN

**Mobile Phase:** A: Water with 0.1 % Formic Acid  
 B: Acetonitrile with 0.1 % Formic Acid

**Gradient:** A/B (90:10) to A/B (10:90) over 6 min

| Step No. | Time(min) | % A | % B |
|----------|-----------|-----|-----|
| 1        | 0         | 90  | 10  |
| 2        | 0.7       | 66  | 34  |
| 3        | 5         | 55  | 45  |
| 4        | 6         | 10  | 90  |

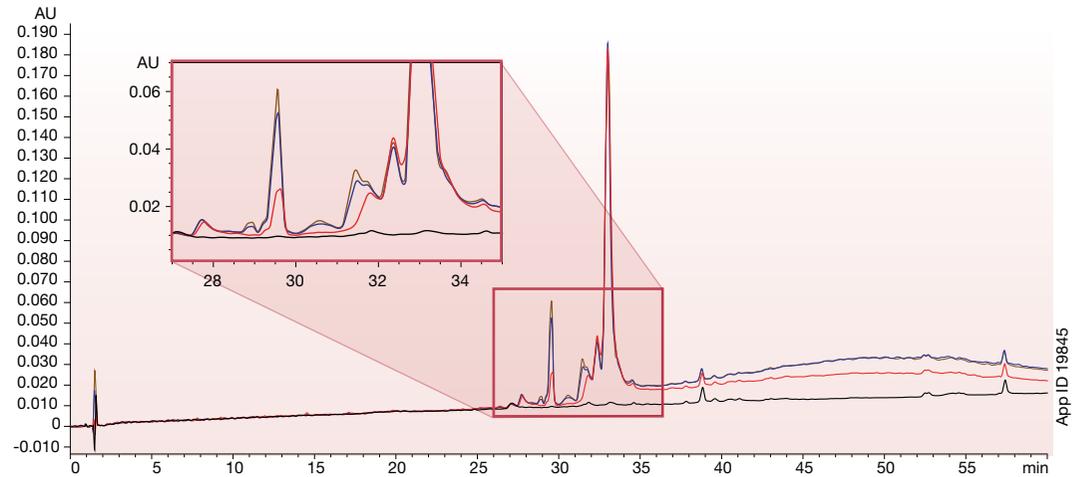
**Flow Rate:** 0.5 mL/min

**Temperature:** 22 °C

**Detection:** UV @ 214 (ambient)

**Sample:** Monoclonal antibody

### Clipped Variants



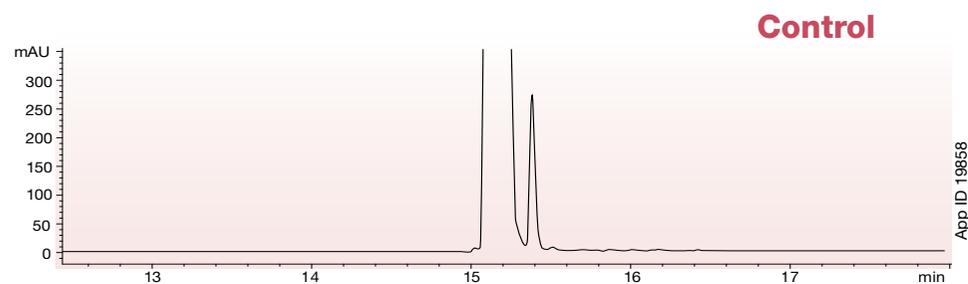
App ID 19845

**Column:** Aeris WIDEPORE 3.6 μm XB-C18  
**Dimensions:** 250 x 4.6 mm  
**Part No.:** 00G-4482-E0  
**Mobile Phase:** A: Water with 0.1 % TFA  
 B: Acetonitrile/IPA (50:50) with 0.1 % TFA  
**Gradient:** A/B (90:10) to A/B (35:65) over 60 min  
**Flow Rate:** 1.0 mL/min  
**Temperature:** 22 °C  
**Injection Volume:** 25 μL  
**Instrument:** Agilent® 1200  
**Detection:** UV @ 214 nm (ambient)  
**Sample:** Proprietary customer monoclonal antibody  
 with clipped variants

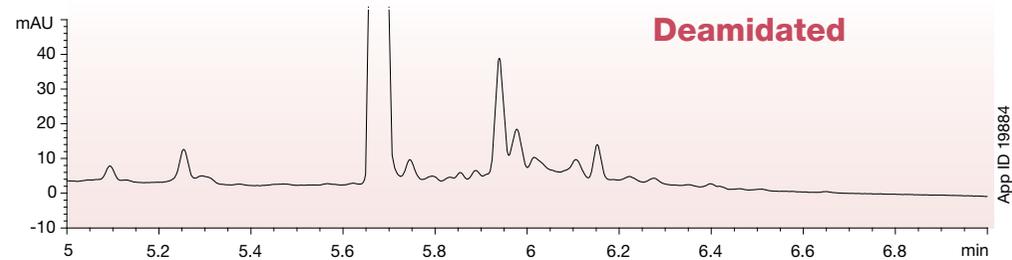
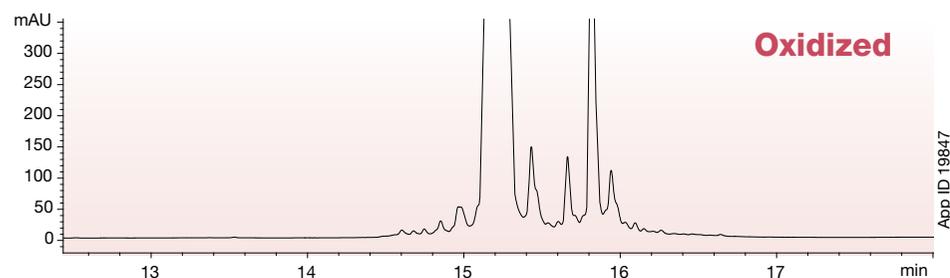
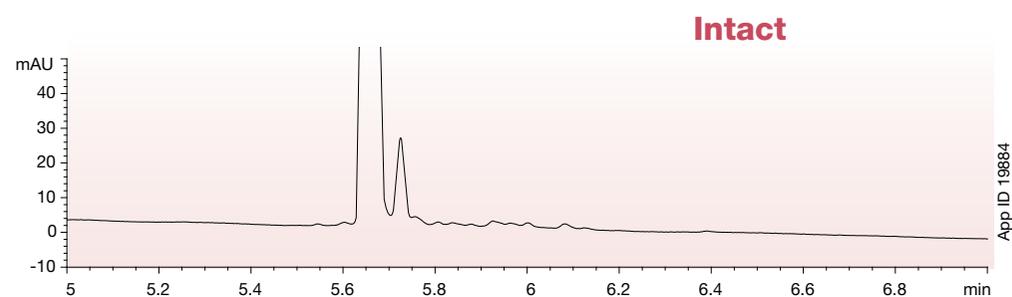
# Applications

## Post-Translational Modification Analysis

### Oxidation



### Deamidation



**Column:** Aeris™ WIDEPORE 3.6µm XB-C18  
**Dimensions:** 100 x 4.6 mm  
**Part No.:** 00D-4482-E0  
**Mobile Phase:** A: Water with 0.1 % TFA  
B: Acetonitrile with 0.1 % TFA  
**Gradient:** A/B (97:3) for 3 min to A/B (15:85) over 45 min

**Flow Rate:** 1.2 mL/min  
**Temperature:** 22°C  
**Injection Volume:** 50 µL  
**Instrument:** Agilent® 1100  
**Detection:** UV @ 214 nm (ambient)  
**Sample:** Insulin oxidized using 3% hydrogen peroxide

**Column:** Aeris WIDEPORE 3.6µm XB-C18  
**Dimensions:** 150 x 4.6 mm  
**Part No.:** 00F-4482-E0  
**Mobile Phase:** A: Water with 0.1 % TFA  
B: Acetonitrile with 0.085 % TFA  
**Gradient:** A/B (90:10) to A/B (35:65) over 10 min

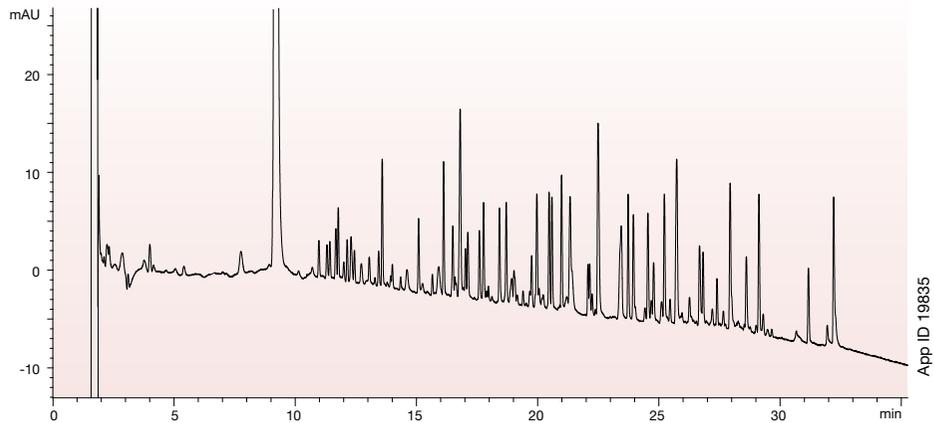
**Flow Rate:** 1.2 mL/min  
**Temperature:** 40°C  
**Injection Volume:** 1 µL  
**Instrument:** Agilent® 1100  
**Detection:** UV @ 214 nm (ambient)  
**Sample:** Proprietary intact insulin 6 kDa deamidated



# Applications

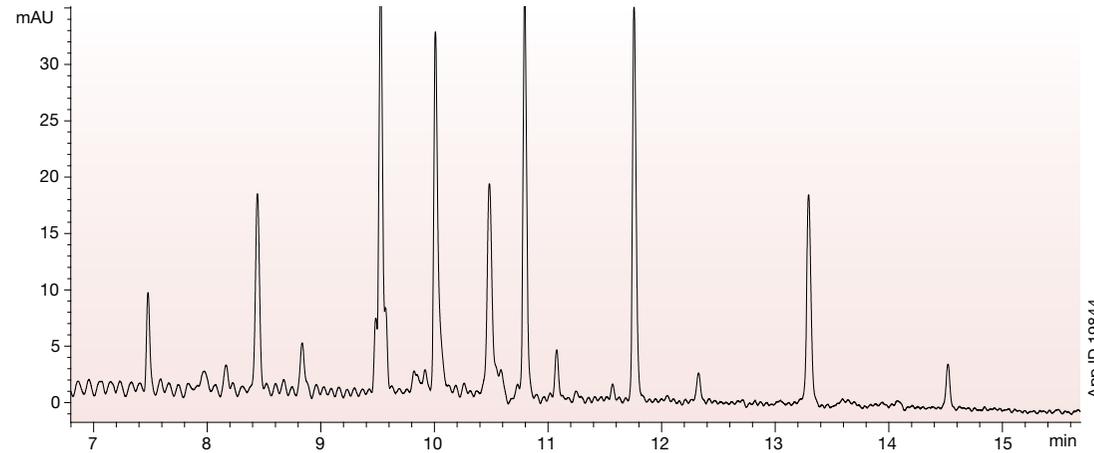
## Digested Protein Analysis

**Bovine Serum Albumin Tryptic Map**



App ID 19835

**Apomyoglobin Digest**



App ID 19844

**Column:** Aeris WIDEPORE 3.6  $\mu$ m XB-C18  
**Dimensions:** 250 x 2.1 mm  
**Part No.:** 00G-4282-AN  
**Mobile Phase:** A: Water with 0.1 % TFA  
 B: Acetonitrile with 0.1 % TFA  
**Gradient:** A/B (97:3) for 3 min to A/B (35:65) over 47 min  
**Flow Rate:** 0.3 mL/min  
**Temperature:** 40  $^{\circ}$ C  
**Injection Volume:** 10  $\mu$ L  
**Instrument:** Agilent<sup>®</sup> 1200SL  
**Detection:** UV @ 214 nm (ambient)  
**Sample:** BSA Tryptic Digest

**Column:** Aeris WIDEPORE 3.6  $\mu$ m XB-C18  
**Dimensions:** 150 x 4.6 mm  
**Part No.:** 00F-4282-E0  
**Mobile Phase:** A: Water with 0.1 % TFA  
 B: Acetonitrile with 0.1 % TFA  
**Gradient:** A/B (97:3) for 3 min to A/B (35:65) over 30 min  
**Flow Rate:** 1.5 mL/min  
**Temperature:** 22  $^{\circ}$ C  
**Injection Volume:** 20  $\mu$ L  
**Instrument:** Agilent<sup>®</sup> 1200  
**Detection:** UV @ 214 nm  
**Sample:** Apomyoglobin Digest

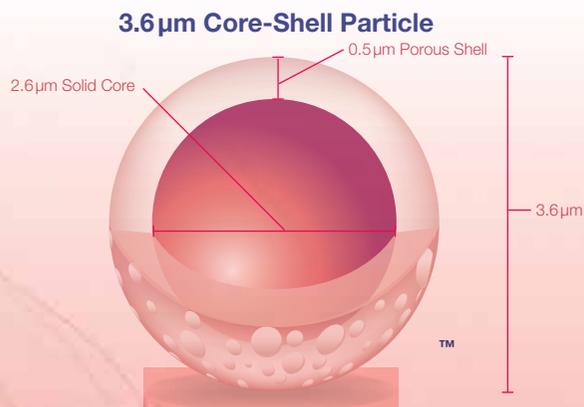
# Aeris™ PEPTIDE Columns

## for Peptide and Peptide Mapping Separations

Based on core-shell particle technology, Aeris PEPTIDE particles are designed with small pores, inert XB-C18 surface chemistry, and three different particle sizes (3.6 μm, 2.6 μm and 1.7 μm) to meet the resolution demands of chromatographers performing complex peptide and peptide map separations on HPLC and/or UHPLC systems.

### Aeris PEPTIDE columns are built for the following:

- Synthetic peptide impurity analysis
- Peptide mapping
- Identifying protein modifications
  - ➔ Glycosylation
  - ➔ Substitution
  - ➔ Truncation
- Analyzing post-translational modifications
  - ➔ Deamidation
  - ➔ Oxidation
  - ➔ Deletions





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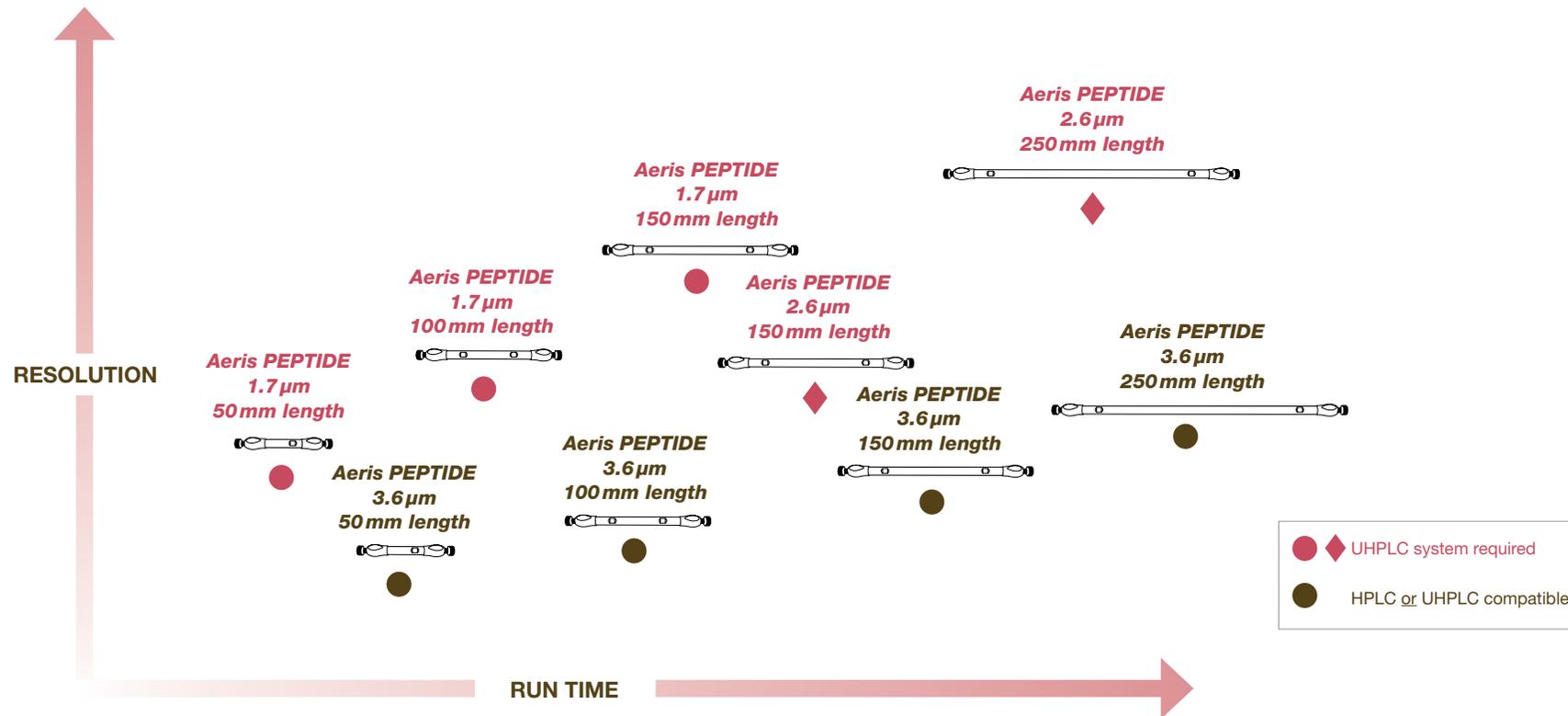
## Aeris PEPTIDE

- p. 28 **Select the Most Suitable Aeris PEPTIDE Column**
- p. 29 **Maximum Performance on UHPLC Systems**
- p. 30 **Ultra-High Resolving Power on HPLC and UHPLC Systems**
- p. 32 **Bundle Aeris PEPTIDE with Aeris WIDEPORE for Detailed Peptide Maps**
- p. 33 **Applications**

“Using the Aeris PEPTIDE column gave us the same resolution separation for cyanobacterial peptides that we could achieve using a smaller particle size column, but with far lower back-pressures. This will allow us to transfer the methods to lower pressure HPLC systems whilst retaining our separation.”

-LARGE PHARMACEUTICAL COMPANY

# Select the Most Suitable Aeris™ PEPTIDE Column to Achieve Your Separation Goals



The family of Aeris PEPTIDE XB-C18 columns is designed to provide versatility for the development of peptide separation methods. Depending on your resolution, throughput goals, and pressure capabilities of your system, you can choose from three particle sizes with unique performance attributes, as well as several column lengths to select the most suitable column for seamless method development and excellent results.

# Maximize Performance on UHPLC Systems with Aeris PEPTIDE 1.7 $\mu\text{m}$ Technology



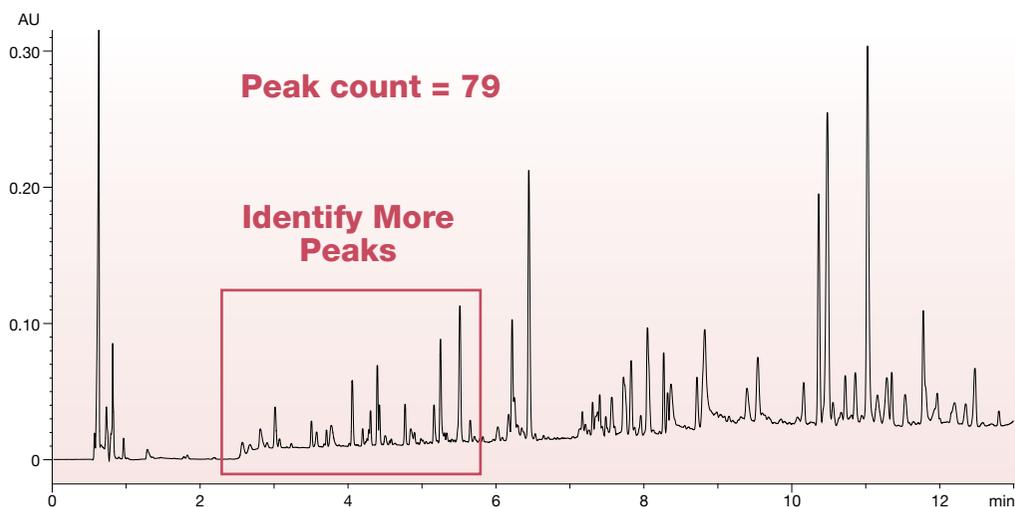
With pressure stability up to 1,000 bar and the high efficiencies brought about by core-shell particle technology, the sub-2  $\mu\text{m}$  Aeris PEPTIDE column produces breakthrough chromatographic performance on UHPLC systems. Use Aeris PEPTIDE 1.7  $\mu\text{m}$  columns to boost the performance of sub-2  $\mu\text{m}$  fully porous peptide mapping methods.

## Increase Peak Count with 1.7 $\mu\text{m}$ Aeris Core-Shell Technology

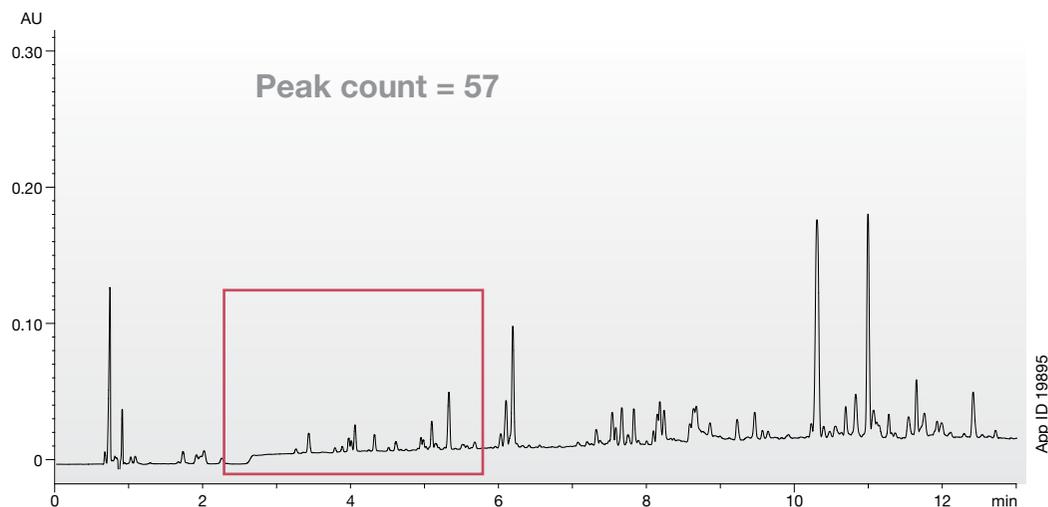
### Conditions for both columns:

**Column:** Aeris PEPTIDE 1.7  $\mu\text{m}$  XB-C18  
ACQUITY® BEH™ 1.7  $\mu\text{m}$  C18  
**Dimensions:** 150 x 2.1 mm  
**Mobile Phase:** A: Water with 0.1 % TFA  
B: Acetonitrile with 0.08 % TFA  
**Gradient:** A/B (97:3) for 1.5 min to A/B (60:40) over 11 min to  
A/B (5:95) over 1 min  
**Flow Rate:** 0.5 mL/min  
**Temperature:** 40 °C  
**Injection Volume:** 5  $\mu\text{L}$   
**Instrument:** Agilent® 1200SL  
**Detection:** UV @ 214 nm (ambient)  
**Sample:** Alpha-Casein Tryptic Digest

### Aeris PEPTIDE 1.7 $\mu\text{m}$ XB-C18



### Waters® ACQUITY® BEH™ 1.7 $\mu\text{m}$ C18

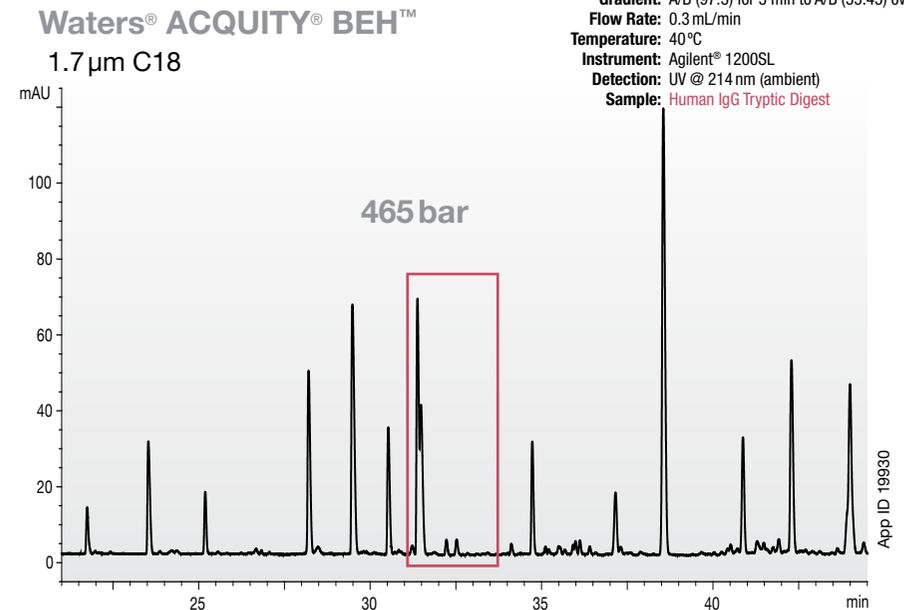
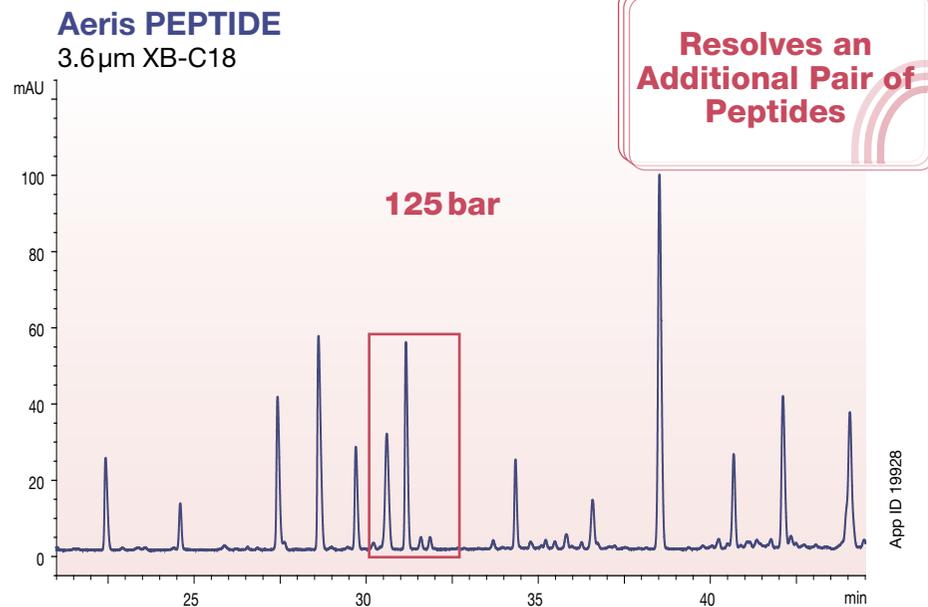


Waters and ACQUITY are registered trademarks, and BEH Technology is a trademark of Waters Corporation. Phenomenex is not affiliated with Waters Corporation. Study was performed using new columns and, to the extent possible, identical experimental conditions were applied. Comparative separations may not be representative of all applications.

# Ultra-High Resolving Power on HPLC and UHPLC Systems with Aeris PEPTIDE 3.6 $\mu\text{m}$ Columns

The Aeris™ PEPTIDE 3.6  $\mu\text{m}$  core shell column was designed with one purpose in mind: to maximize the separation of large numbers of peptides on any HPLC or UHPLC system. Because core-shell particles remove the backpressure constraints of HPLC or UHPLC systems, chromatographers can **achieve the ultra-high performance of similar length sub-2  $\mu\text{m}$  columns at a fraction of the backpressure.**

**Conditions for both columns:**  
**Column:** Aeris PEPTIDE 3.6  $\mu\text{m}$  XB-C18  
 ACQUITY® BEH™ 1.7  $\mu\text{m}$  C18  
**Dimensions:** 150 x 2.1 mm  
**Mobile Phase:** A: Water with 0.1 % TFA  
 B: Acetonitrile with 0.1 % TFA  
**Gradient:** A/B (97:3) for 5 min to A/B (55:45) over 55 min  
**Flow Rate:** 0.3 mL/min  
**Temperature:** 40 °C  
**Instrument:** Agilent® 1200SL  
**Detection:** UV @ 214 nm (ambient)  
**Sample:** Human IgG Tryptic Digest

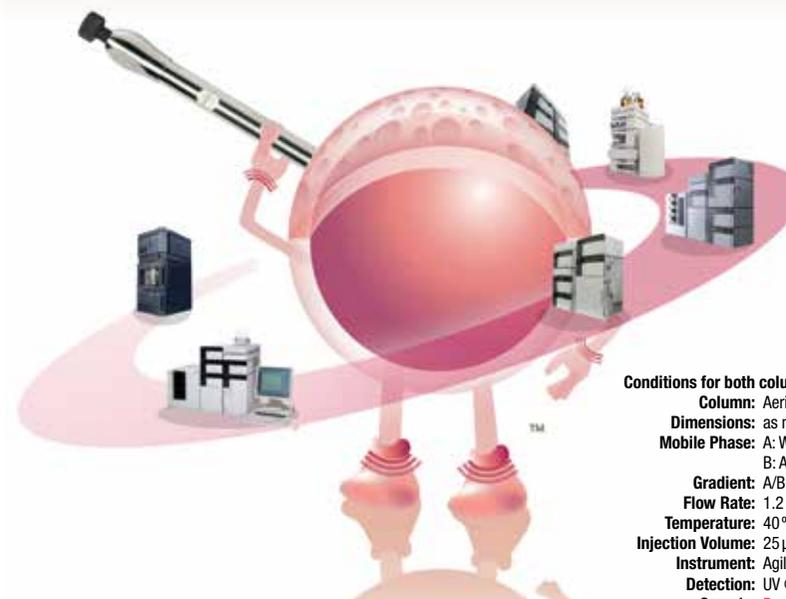


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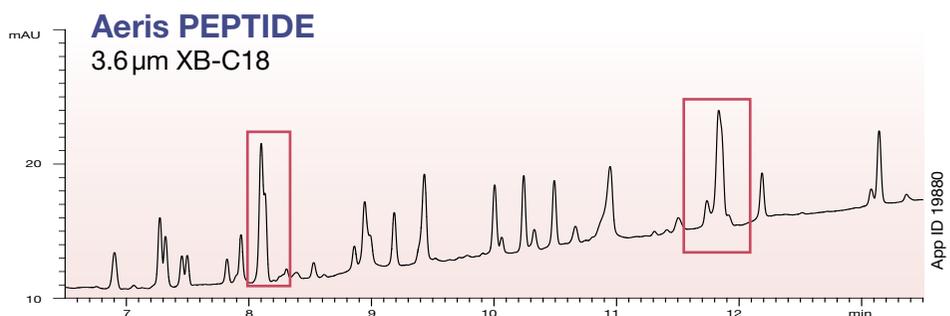
**Use longer (or coupled) 3.6 μm columns on UHPLC and HPLC systems to resolve critical peaks**

For applications like peptide separations and peptide mapping where resolution is the primary goal, the lower backpressure of Aeris PEPTIDE 3.6 μm core-shell columns allow one to use longer columns for higher resolving power resulting in increased separation of closely eluting peptides.



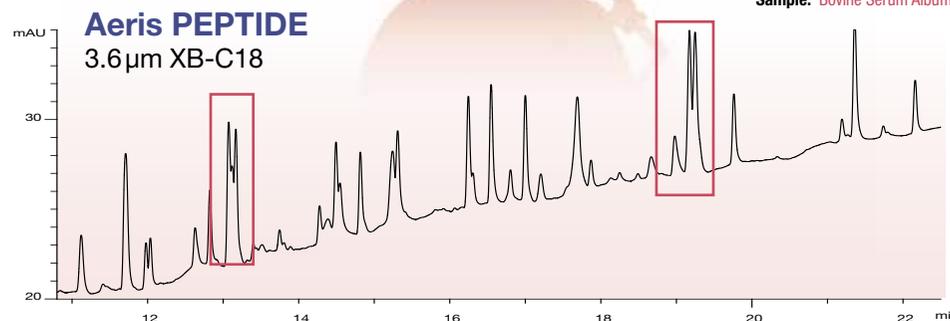
**Conditions for both columns:**  
**Column:** Aeris PEPTIDE 3.6 μm XB-C18  
**Dimensions:** as noted  
**Mobile Phase:** A: Water with 0.1 % Formic Acid  
 B: Acetonitrile with 0.1 % Formic Acid  
**Gradient:** A/B (97:3) for 3 min to A/B (35:65) over 30 min  
**Flow Rate:** 1.2 mL/min  
**Temperature:** 40 °C  
**Injection Volume:** 25 μL  
**Instrument:** Agilent® 1200  
**Detection:** UV @ 214 nm (ambient)  
**Sample:** Bovine Serum Albumin (BSA) Tryptic Digest

**Utilize Long Columns to Maximize Separation Power**



**150 x 4.6 mm**

**140 bar**



**250 x 4.6 mm**

**200 bar**

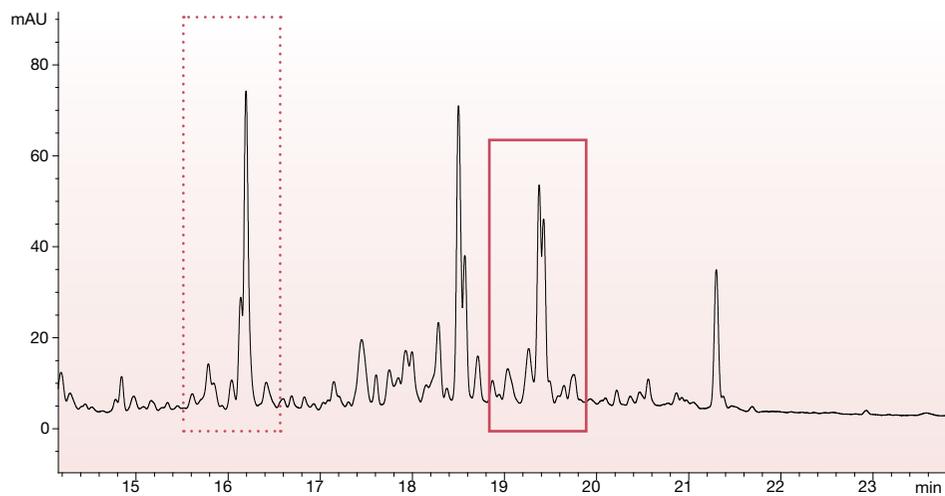
**Minimal Increase in Backpressure**

# Bundle Aeris™ PEPTIDE with Aeris WIDEPORE for Detailed Peptide Maps

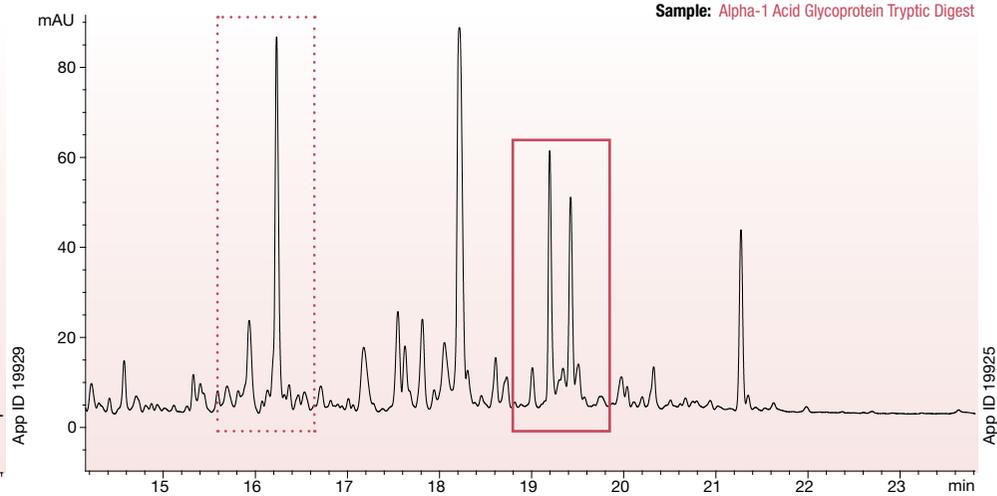
Aeris PEPTIDE 3.6µm XB-C18 and Aeris WIDEPORE 3.6µm XB-C18 are a “must-have” pair for chromatographers who analyze complex peptide mixtures. Because each has a unique pore size and surface area, they exhibit different selectivity. Protein chemists can take advantage of this diversity to achieve the critical resolution of target peptides in various regions of the map, thus simplifying their method development.

## Utilize Differences in Small and Large Pore Size Selectivity for Optimal Resolution

### Aeris PEPTIDE 3.6µm XB-C18



### Aeris WIDEPORE 3.6µm XB-C18



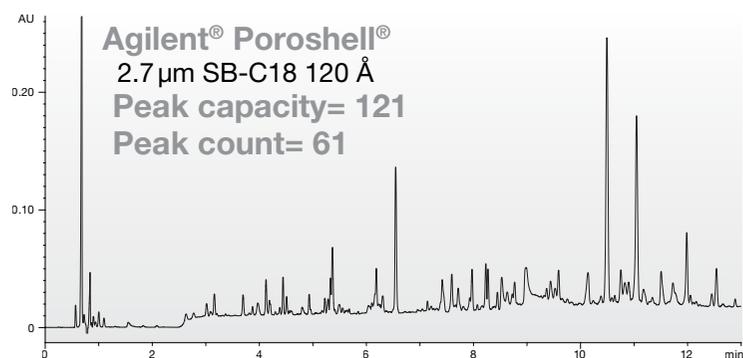
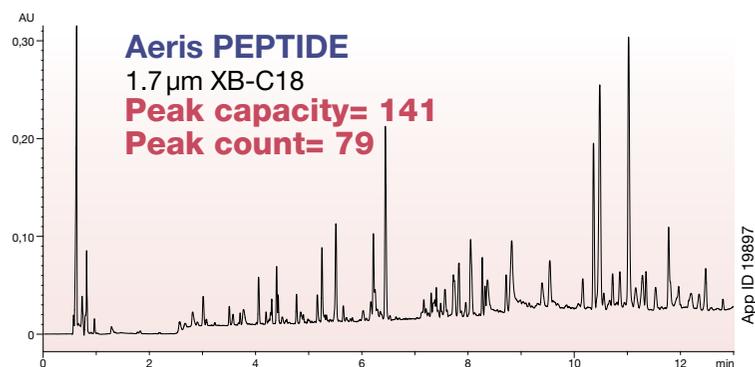
**Conditions for both columns:**  
**Column:** Aeris PEPTIDE 3.6µm XB-C18  
Aeris WIDEPORE 3.6µm XB-C18  
**Dimensions:** 150 x 4.6 mm  
**Part Nos.:** 00F-4507-E0  
00F-4482-E0  
**Mobile Phase:** A: Water with 0.1 % TFA  
B: Acetonitrile with 0.1 % TFA  
**Gradient:** A/B (97:3) for 3 min to A/B (35:65) over 30 min  
**Flow Rate:** 1.5 mL/min  
**Temperature:** 40°C  
**Instrument:** Agilent® 1200SL  
**Detection:** UV @ 214 nm (ambient)  
**Sample:** Alpha-1 Acid Glycoprotein Tryptic Digest



# Applications

## Peptide Mapping on Core-Shell Technologies

### Aeris PEPTIDE vs. Other Core-Shell Columns



**Conditions same for all columns:**

**Columns:** Aeris PEPTIDE 1.7 μm XB-C18  
 Poroshell® 2.7 μm SB-C18 120 Å  
 Ascentis® Express Peptide 2.7 μm C18

**Dimensions:** 150 x 2.1 mm

**Mobile Phase:** A: Water with 0.1 % Formic Acid  
 B: Acetonitrile with 0.08 % Formic Acid

**Gradient:** A/B (97:3) for 1.5 min to A/B (60:40)  
 over 11 min to A/B (5:95) over 1 min

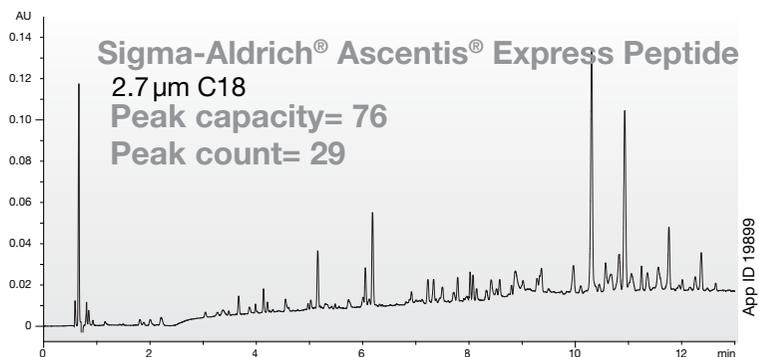
**Flow Rate:** 0.5 mL/min

**Temperature:** 40 °C

**Instrument:** Agilent® 1200SL

**Detection:** UV @ 214 nm (ambient)

**Sample:** Alpha-Casein Tryptic Digest



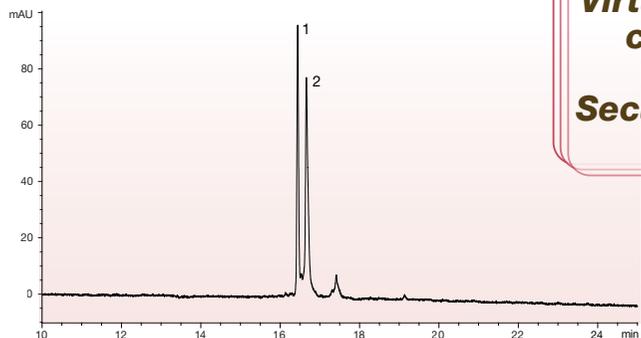
Agilent and Poroshell are registered trademarks of Agilent Technologies, Inc. Ascentis Express Peptide is a registered trademark of Sigma-Aldrich Biotechnology. Phenomenex is not affiliated with Agilent Technologies, Inc or Sigma-Aldrich Biotechnology. Comparative separations may not be representative of all applications.

# Extend the Lifetime of your Aeris Core-Shell Columns with SecurityGuard ULTRA

The SecurityGuard ULTRA guard cartridge system protects Aeris core-shell columns from damaging chemical contaminants, protein adsorption, and microparticulates. This innovative and easy-to-use column protection system will not alter chromatography or contribute to extra dead volume and is pressure rated up to 20,000 psi for UHPLC systems.



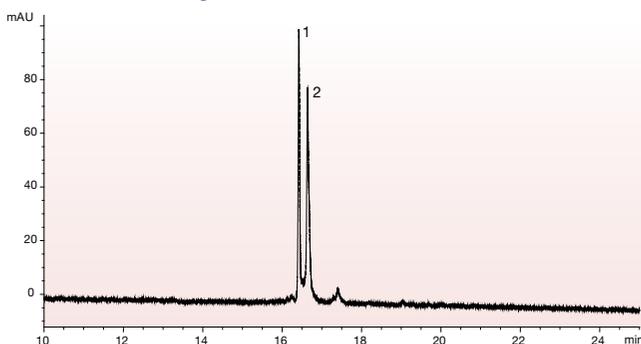
**With SecurityGuard ULTRA**



**Virtually no change in chromatography when using SecurityGuard ULTRA!**

App ID 19924

**Without SecurityGuard ULTRA**



App ID 19923

Conditions same for both separations:

**Columns:** Aeris WIDEPORE 3.6 µm XB-C18

**Dimensions:** 150 x 4.6 mm

**Mobile Phase:** A: Water with 0.1% TFA

B: Acetonitrile with 0.085% TFA

**Gradient:** A/B (97:3) for 3 min to A/B (35:65) over 30 min

**Flow Rate:** 1.2 mL/min

**Temperature:** 40 °C

**Instrument:** Agilent® 1200

**Detection:** UV @ 214 nm (ambient)

**Sample:** 1. RNase A

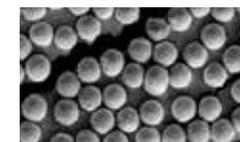
2. Reduced RNase A

**With SecurityGuard ULTRA**



**Inlet Frit**

**Column Media**

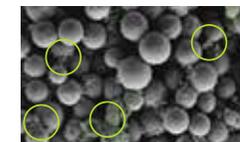


**Without SecurityGuard ULTRA**



**Inlet Frit**

**Column Media**



**Contaminant and particle buildup**

**guarantee**

If SecurityGuard ULTRA cartridge protection system does not perform as well or better than your current guard cartridge system of similar phase and dimensions, return the cartridge with comparative data within 45 days for a FULL REFUND.

Ordering information on page 35



# Ordering Information



| Aeris PEPTIDE 1.7 µm Minibore Columns (mm) |             |             |             | SecurityGuard<br>ULTRA Cartridges* |
|--|-------------|-------------|-------------|------------------------------------|
| Phases                                     | 50 x 2.1    | 100 x 2.1   | 150 x 2.1   | 3/pk                               |
| XB-C18                                     | 00B-4506-AN | 00D-4506-AN | 00F-4506-AN | AJO-8948                           |

| Aeris PEPTIDE 2.6 µm Minibore Columns (mm) |             |             | SecurityGuard™<br>ULTRA Cartridges* |
|--|-------------|-------------|-------------------------------------|
| Phases                                     | 150 x 2.1   | 250 x 2.1   | 3/pk                                |
| XB-C18                                     | 00F-4505-AN | 00G-4505-AN | AJO-8948                            |

| Aeris PEPTIDE 2.6 µm Analytical Columns (mm) |             |             | SecurityGuard<br>ULTRA Cartridges* |
|--|-------------|-------------|------------------------------------|
| Phases                                       | 150 x 4.6   | 250 x 4.6   | 3/pk                               |
| XB-C18                                       | 00F-4505-E0 | 00G-4505-E0 | AJO-8946                           |

| Aeris PEPTIDE 3.6 µm Minibore Columns (mm) |             |             |             |             | SecurityGuard<br>ULTRA Cartridges* |
|--|-------------|-------------|-------------|-------------|------------------------------------|
| Phases                                     | 50 x 2.1    | 100 x 2.1   | 150 x 2.1   | 250 x 2.1   | 3/pk                               |
| XB-C18                                     | 00B-4507-AN | 00D-4507-AN | 00F-4507-AN | 00G-4507-AN | AJO-8948                           |

| Aeris PEPTIDE 3.6 µm Analytical Columns (mm) |             |             |             |             | SecurityGuard<br>ULTRA Cartridges* |
|--|-------------|-------------|-------------|-------------|------------------------------------|
| Phases                                       | 50 x 4.6    | 100 x 4.6   | 150 x 4.6   | 250 x 4.6   | 3/pk                               |
| XB-C18                                       | 00B-4507-E0 | 00D-4507-E0 | 00F-4507-E0 | 00G-4507-E0 | AJO-8946                           |

## Material Characteristics

| Packing Material | Total Particle Size (µm) | Porous Shell (µm) | Core Size (µm) | pH Stability | Temp Stability | Pressure Stability |
|------------------|--------------------------|-------------------|----------------|--------------|----------------|--------------------|
| Aeris WIDEPORE   | 3.6                      | 0.2               | 3.2            | 1.5 - 9      | 90 °C          | 600 bar            |
| Aeris PEPTIDE    | 1.7                      | 0.22              | 1.25           | 1.5 - 9      | 90 °C          | 1000 bar           |
| Aeris PEPTIDE    | 2.6                      | 0.35              | 1.9            | 1.5 - 9      | 90 °C          | 1000 bar           |
| Aeris PEPTIDE    | 3.6                      | 0.5               | 2.6            | 1.5 - 9      | 90 °C          | 600 bar            |

| Aeris WIDEPORE 3.6 µm Minibore Columns (mm) |             |             |             |             | SecurityGuard™<br>ULTRA Cartridges* |
|---|-------------|-------------|-------------|-------------|-------------------------------------|
| Phases                                      | 50 x 2.1    | 100 x 2.1   | 150 x 2.1   | 250 x 2.1   | 3/pk                                |
| XB-C18                                      | 00B-4482-AN | 00D-4482-AN | 00F-4482-AN | 00G-4482-AN | AJO-8783                            |
| XB-C8                                       | 00B-4481-AN | 00D-4481-AN | 00F-4481-AN | 00G-4481-AN | AJO-8785                            |
| C4  | 00B-4486-AN | 00D-4486-AN | 00F-4486-AN | 00G-4486-AN | AJO-8899                            |

| Aeris WIDEPORE 3.6 µm Analytical Columns (mm) |             |             |             | SecurityGuard<br>ULTRA Cartridges* |
|---|-------------|-------------|-------------|------------------------------------|
| Phases  | 100 x 4.6   | 150 x 4.6   | 250 x 4.6   | 3/pk                               |
| XB-C18  | 00D-4482-E0 | 00F-4482-E0 | 00G-4482-E0 | AJO-8769                           |
| XB-C8   | 00D-4481-E0 | 00F-4481-E0 | 00G-4481-E0 | AJO-8771                           |
| C4  | 00D-4486-E0 | 00F-4486-E0 | 00G-4486-E0 | AJO-8901                           |

\*SecurityGuard ULTRA cartridges require holder part number. AJO-9000

| SecurityGuard ULTRA Cartridge Holder* (for 2.1 to 4.6 mm ID columns) |    |          |
|--|----|----------|
| SecurityGuard ULTRA Guard Cartridge Holder                           | ea | Price    |
|  |    | AJO-9000 |

## Ordering Information

| Core-Shell Performance Enhancement Kit                 |  |                     |       |
|--|--|---------------------|-------|
| Part No.   | Description  | Unit                | Price |
| AQO-8892   | Core-Shell Performance Enhancement Kit, Includes: PEEKsil™ Tubing, Fittings and Tool | ea                  |       |
| <b>Kit AQO-8892 includes the following components:</b> |  | <b>Kit Quantity</b> |       |
|  | PEEKsil Tubing 0.100 mm ID x 1/16 in. OD x 20 cm L, Red                              | 2/pk                |       |
|  | PEEKsil Tubing 0.100 mm ID x 1/16 in. OD x 10 cm L, Red                              | ea                  |       |
|  | Sure-Lok™ High Pressure PEEK 1-Pc Nut, 10-32, for 1/16 in. Tubing                    | 10/pk               |       |
|  | Sure-Lok Fitting Tightening Tool, Aluminum   | ea                  |       |



If you are not completely satisfied with your Aeris core-shell columns, return the column with comparative data within 45 days for a FULL REFUND.

**Australia**

t: 02-9428-6444  
 f: 02-9428-6445  
 auinfo@phenomenex.com

**Austria**

t: 01-319-1301  
 f: 01-319-1300  
 anfrage@phenomenex.com

**Belgium**

t: 02 503 4015 (French)  
 t: 02 511 8666 (Dutch)  
 f: +31 (0)30-2383749  
 beinfo@phenomenex.com

**Canada**

t: (800) 543-3681  
 f: (310) 328-7768  
 info@phenomenex.com

**Denmark**

t: 4824 8048  
 f: +45 4810 6265  
 nordicinfo@phenomenex.com

**Finland**

t: 09 4789 0063  
 f: +45 4810 6265  
 nordicinfo@phenomenex.com

**France**

t: 01 30 09 21 10  
 f: 01 30 09 21 11  
 franceinfo@phenomenex.com

**Germany**

t: 06021-58830-0  
 f: 06021-58830-11  
 anfrage@phenomenex.com

**India**

t: 040-3012 2400  
 f: 040-3012 2411  
 indiainfo@phenomenex.com

**Ireland**

t: 01 247 5405  
 f: +44 1625-501796  
 eireinfo@phenomenex.com

**Italy**

t: 051 6327511  
 f: 051 6327555  
 italiainfo@phenomenex.com

**Luxembourg**

t: +31 (0)30-2418700  
 f: +31 (0)30-2383749  
 nlinfo@phenomenex.com

**Mexico**

t: 001-800-844-5226  
 f: 001-310-328-7768  
 tecnicomx@phenomenex.com

**The Netherlands**

t: 030-2418700  
 f: 030-2383749  
 nlinfo@phenomenex.com

**New Zealand**

t: 09-4780951  
 f: 09-4780952  
 nzinfo@phenomenex.com

**Norway**

t: 810 02 005  
 f: +45 4810 6265  
 nordicinfo@phenomenex.com

**Puerto Rico**

t: (800) 541-HPLC  
 f: (310) 328-7768  
 info@phenomenex.com

**Sweden**

t: 08 611 6950  
 f: +45 4810 6265  
 nordicinfo@phenomenex.com

**United Kingdom**

t: 01625-501367  
 f: 01625-501796  
 ukinfo@phenomenex.com

**United States**

t: (310) 212-0555  
 f: (310) 328-7768  
 info@phenomenex.com

**All other countries:  
Corporate Office USA** 

t: (310) 212-0555  
 f: (310) 328-7768  
 info@phenomenex.com

Core-Shell Technology  
for Proteins and Peptides

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