

BIOshell[™] Fused-Core[®] Columns

For Faster Peptide and Protein Reversed-Phase Liquid Chromatography



BIOshell[™] U/HPLC Columns

Improve Your Separation
without Increasing Pressure

Maintain Quality while
Improving Throughput

Opportunity for Faster or Better Separations

For HPLC and UHPLC Systems

BIOshell™ Fused-Core® Columns

Opportunity For Faster or Better Peptide and Protein Separations

BIOshell columns are the most recent innovation in Fused-Core particle technology: high efficiency reversed-phase columns for protein and peptide separations. BIOshell columns can be operated in HPLC or UHPLC instrumentation equipped with a mass spectrometer or any other detector. BIOshell A160 Peptide columns are packed with either 2.7 or 5 micron particles containing 160 Å pores and are available with a C18 alkyl or alkyl cyano (CN) bonded phase functionality. BIOshell A400 Protein columns are packed with 3.4 micron particles containing 400 Å pores and are bonded with C4 alkyl functional groups.

Particle Characteristics of BIOshell Fused-Core Columns

BIOshell Columns	Particle Size (µm)	Core Size (µm)	Shell Thickness (µm)	Pore Size (Å)	SBET (m ² /g)	Capacity vs. Porous*
2.7 µm A160 Peptide C18	2.7	1.7	0.5	160	80	75%
2.7 µm A160 Peptide CN	2.7	1.7	0.5	160	80	75%
5 µm A160 Peptide C18	4.7+	3.5	0.6	160	80	59%
5 µm A160 Peptide CN	4.7+	3.5	0.6	160	80	59%
3.4 µm A400 Protein C4	3.4	3.0	0.2	400	15	31%

+ Nominal particle size is 5 micron

* Calculated capacity based on value of core diameter and shell thickness

Bonded Phase and Operational Characteristics of BIOshell Fused-Core Columns

BIOshell Columns	Bonded Phase Ligand	End Cap	Max. Temp. (°C)*	pH Range	Pmax (bar)	Frit Porosity (µm)
2.7 µm A160 Peptide C18	di-isobutyl-octadecylsilane	No	100	1 - 8	600	2
2.7 µm A160 Peptide CN	di-isopropyl-cyanopropylsilane	Yes	80	1 - 8	600	2
5 µm A160 Peptide C18	di-isobutyl-octadecylsilane	No	100	1 - 8	600	2
5 µm A160 Peptide CN	di-isopropyl-cyanopropylsilane	Yes	90	1 - 8	600	2
3.4 µm A400 Protein C4	dimethylbutylsilane	Yes	90	2 - 9	600	2

Temperature at which each bonded phase type was tested for long term physical and chemical stability.

Features and Benefits

- The efficiency of core-shell particles is about 40% higher than that of fully porous particles of the same size.
- Dialkyl silane reagents provide extra bonded phase stability.
- High operating temperatures increase throughput and improve peak shape and efficiency of strongly hydrophobic peptides and proteins.
- Each column type was tested up to 600 bar pressure to allow operation at high flow rate.
- Narrow particle size distributions allow the use of 2 micron porosity frits, even for 2.7 micron particles*.
- BIOshell Fused-Core columns are rugged, robust and reliable.

*While the use of 2 micron porosity frits means that BIOshell Fused-Core columns do not plug easily, the user is still advised to clean samples before injection with a 0.2 micron syringe filter.

Selected BIOshell™ Applications

The chromatogram in **Figure 1** shows the analysis of IgG2-B subtype monoclonal antibody following solubilization and reduction of the sulfide bonds with 100 mM dithiothreitol (DTT) in 8 M guanidine HCl at 50 °C for 35 minutes.

The main components in **Figure 1** are the light (LC) and heavy (HC) chains of IgG2-B. The inset clearly shows several expected variants near the heavy chain peak.

As shown in **Figure 2**, BIOshell A400 Protein C4 columns can be operated at 90 °C, at which temperature peak widths of IgG fragments are narrower and peak shape is better than on a competitive column packed with fully porous 1.7 micron particles, operated at the highest temperature recommended by its manufacturer.

It is well-known that columns packed with core-shell particles provide higher efficiency than columns packed with fully porous particles of the same size. This is apparent in **Figure 2**, but can also be confirmed by calculating the peak capacity for a tryptic digest sample. Injecting a mixture of 5 different tryptic digests on a 2.7 micron BIOshell A160 Peptide C18 column resulted in the chromatogram shown in **Figure 3**. A peak capacity of 340 was calculated based on a gradient time of 41.3 minutes. When the same sample was analyzed on a column packed with 5 micron C18-bonded porous particles, the peak capacity was 240.

The examples shown here demonstrate that BIOshell Fused-Core columns provide very high efficiency separations for peptide and protein samples that are routinely analyzed in academic, biopharmaceutical, clinical and diagnostic laboratories. These new column types provide the scientist with the option of either using the extra efficiency to reduce analysis time or obtain better quality data by separating more components per unit time.

Figure 1. High Temperature Analysis of IgG2-B Antibody Fragments on BIOshell A400 Protein C4 Column

column: BIOshell A400 Protein C4, 10 cm x 2.1 mm I.D., 3.4 µm (66825-U)
 mobile phase: A: water:0.1% TFA, B: acetonitrile:water:0.1% TFA, 80:20
 gradient: 33-40% B in 10 min
 flow rate: 0.25 mL/min
 temp: 80 °C
 injection vol: 1 µL
 detection: 280 nm
 sample: 0.5 mg/mL IgG2-B treated with 100 mM DDT in 8 M guanidine HCl at 50 °C for 35 min
 instrument: Shimadzu® Nexera®

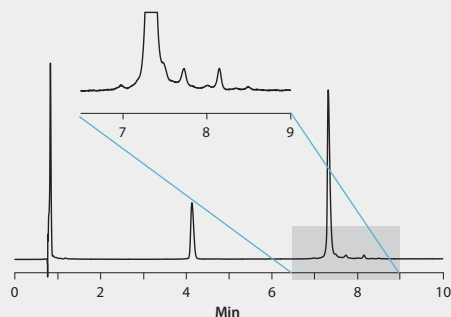
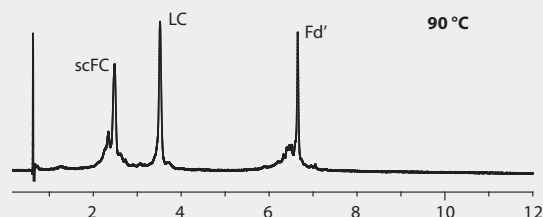


Figure 2. Analysis of Antibody Fragments on Wide Pore Reversed Phase Columns Operated at Maximum Recommended Temperature.

column: as indicated, 10 cm x 2.1 mm
 mobile phase A: 80:20, (water, 0.1% TFA) : (acetonitrile, 0.1% TFA)
 mobile phase B: 50:50, (water, 0.1% TFA) : (acetonitrile, 0.1% TFA)
 gradient: 30 to 70% B in 12 min
 flow: 0.3 mL/min
 column temp.: as indicated
 detection: UV, 215 nm
 injection: 1 µL, after sample diluted in mobile phase A

BIOshell A400 Protein C4, 3.4 µm



Acquity UPLC BEH300 C4, 1.7 µm

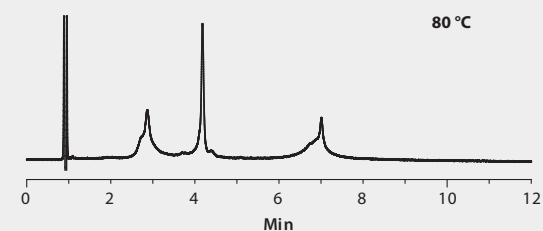
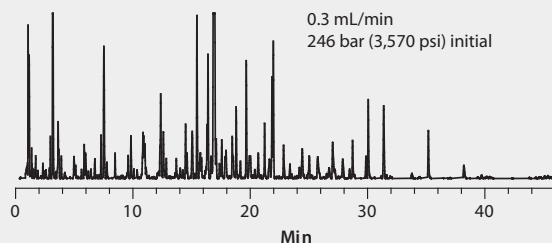


Figure 3. Peptide Digest Mixture on BIOshell A160 Peptide C18 Column

columns: BIOshell A160 Peptide C18, 15 cm x 2.1 mm I.D., 2.7 µm (66905-U)
 mobile phase A: 0.1 % formic acid
 mobile phase B: 25:75, (0.4 % formic acid):acetonitrile
 gradient: 6-70% B/40 min
 flow rate: 0.3 mL/min
 column temp.: 35 °C
 det.: ESI(+)-TOF
 injection: 2 µL
 sample: 10 pmol/µL



Acknowledgement: Stephanie A. Schuster and Robert E. Moran from Applied Materials Technologies, Inc. contributed Figure 1.

Featured Products

Pore Size	Particle Size	I.D. (mm)	L (cm)	C4	C18	CN
BIOshell Fused-Core Peptide and Protein Columns						
400 Å	3.4 µm	2.1	5	66824-U	—	—
400 Å	3.4 µm	2.1	10	66825-U	—	—
400 Å	3.4 µm	2.1	15	66826-U	—	—
400 Å	3.4 µm	4.6	5	66827-U	—	—
400 Å	3.4 µm	4.6	10	66828-U	—	—
400 Å	3.4 µm	4.6	15	66829-U	—	—
160 Å	2.7 µm	2.1	3	—	66901-U	66965-U
160 Å	2.7 µm	2.1	5	—	66902-U	66966-U
160 Å	2.7 µm	2.1	7.5	—	66903-U	66967-U
160 Å	2.7 µm	2.1	10	—	66904-U	66968-U
160 Å	2.7 µm	2.1	15	—	66905-U	66969-U
160 Å	2.7 µm	3.0	3	—	66906-U	66970-U
160 Å	2.7 µm	3.0	5	—	66907-U	66971-U
160 Å	2.7 µm	3.0	10	—	66908-U	66972-U
160 Å	2.7 µm	3.0	15	—	66909-U	66973-U
160 Å	2.7 µm	4.6	5	—	66913-U	66974-U
160 Å	2.7 µm	4.6	10	—	66915-U	66975-U
160 Å	2.7 µm	4.6	15	—	66917-U	66976-U
160 Å	5 µm	2.1	3	—	67001-U	67061-U
160 Å	5 µm	2.1	5	—	67002-U	67062-U
160 Å	5 µm	2.1	7.5	—	67003-U	67063-U

Pore Size	Particle Size	I.D. (mm)	L (cm)	C4	C18	CN
160 Å	5 µm	2.1	10	—	67004-U	67064-U
160 Å	5 µm	2.1	15	—	67006-U	67065-U
160 Å	5 µm	3.0	3	—	67007-U	67066-U
160 Å	5 µm	3.0	5	—	67008-U	67067-U
160 Å	5 µm	3.0	10	—	67011-U	67068-U
160 Å	5 µm	3.0	15	—	67012-U	67069-U
160 Å	5 µm	4.6	5	—	67013-U	67071-U
160 Å	5 µm	4.6	10	—	67014-U	67080-U
160 Å	5 µm	4.6	15	—	67015-U	67081-U
BIOshell Fused-Core Peptide and Protein Guard Columns						
400 Å	3.4 µm	2.1	1	66830-U	—	—
400 Å	3.4 µm	4.6	1	66831-U	—	—
160 Å	2.7 µm	2.1	1	—	66918-U	66977-U
160 Å	2.7 µm	3.0	1	—	66919-U	66978-U
160 Å	2.7 µm	4.6	1	—	66921-U	66979-U
160 Å	5 µm	2.1	1	—	67016-U	67082-U
160 Å	5 µm	3.0	1	—	67017-U	67083-U
160 Å	5 µm	4.6	1	—	67018-U	67084-U

Related Product

Description	Cat. No.
BIOshell Guard Cartridge Holder	66841-U

Other U/HPLC Columns for Proteins and Peptides Available from Sigma-Aldrich

Supplier, Manufacturer	dp (µm)	Matrix	Size Exclusion	Ion Exchange	Reversed Phase	HILIC	HIC	Affinity
GE Life Sciences	8, 13	dextran-agarose	Superdex™					
	>10 (dry)	dextran	Desalting					
	3, 10	PS/DVB		MiniBeads™, MonoBeads™				
	15	PS/DVB		SOURCE™	SOURCE™		SOURCE™	
Sepax Technologies	3	silica	Zenix®, Zenix-C					
	5	silica	SRT®, SRT-C					
	1.7, 3, 5, 10	PS/DVB		Proteomix®				
	1.7, 3, 5, 10	PS/DVB		Antibodix®				
Supelco/ Sigma-Aldrich	2.7, 3.4, 5	silica			BIOshell			
	3, 5, 10	silica			Discovery® BIO			
	5	methacrylate						
Tosoh Bioscience®: TSKgel®	3, 4, 5, 10	silica	UltraSW, SuperSW, SWxl, SW	Discovery BIO PolyMA				
	2.5, 5, 10	methacrylate		NPR, STAT, 5PW				
	2.5, 10	methacrylate			NPR, 4PW, 5PW			
	3	silica			Peptide C4			
	3, 5	silica				Amide-80		
	2.5, 10	methacrylate					NPR, 5PW	
	10	methacrylate						5PW

For more information, visit sigma-aldrich.com/bio-hplc

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