

SEE YOUR  
**PEPTIDES**  
**IN A NEW LIGHT**

Enhanced Selectivity With the HALO® 160Å Phenyl-Hexyl Column

**HALO**®

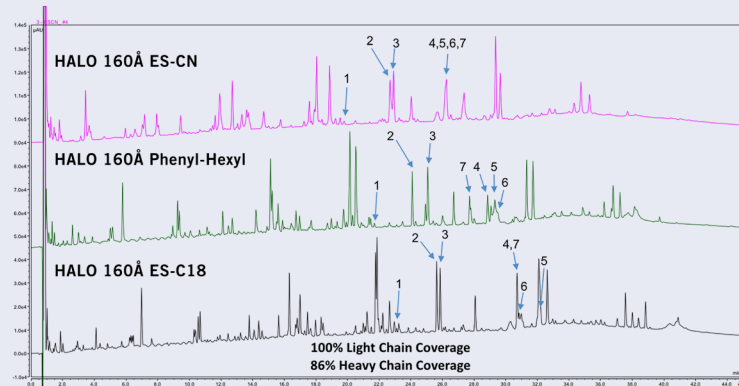
 advancedmaterialstechnology

## INTRODUCING HALO® 160Å PHENYL-HEXYL

HALO® 160Å Phenyl-Hexyl columns are specifically designed to offer alternate selectivity to HALO 160Å ES-C18 and HALO 160Å ES-CN for separations of peptides and tryptic digests. The Fused-Core® particle design with a total particle size of 2.7 µm and 0.5 µm shell with 160Å pores enables high resolution at elevated flow rates. Additionally, HALO 160Å Phenyl-Hexyl columns have similar efficiency to sub-2-micron columns without the inconvenience of higher pressure.

### UNIQUE SELECTIVITY

Figure 1. The unique selectivity of HALO 160Å Phenyl-Hexyl enables different resolutions for tryptic digest fragments compared to HALO 160Å ES-C18 and HALO 160Å ES-CN.



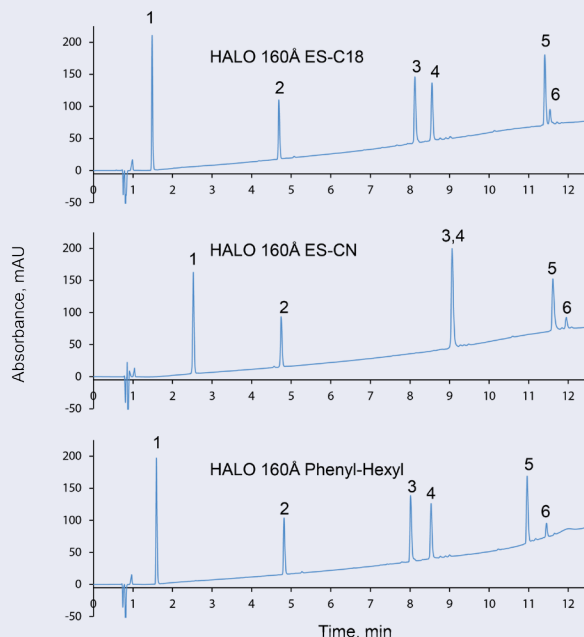
#### PEAK IDENTITIES:

- |                           |                                   |
|---------------------------|-----------------------------------|
| 1. FTISADTSKNTAYLQMNSLR   | 5. SGTASVVCLLNNFYPR               |
| 2. LScAASGFNIKDTYIHWVR    | 6. ScDKTHTcPPcPAPELLGGPSVFLFPPKPK |
| 3. GFYPSDIAVEWESNGQPENNYK | 7. VVSLTVLHQDWLNGKEYK             |
| 4. LLIYSASFLYSGVPSR       |                                   |

#### TEST CONDITIONS:

Column: 2.1 x 100 mm  
 Top: HALO 160Å ES-CN, 2.7 µm  
 Middle: HALO 160Å Phenyl-Hexyl, 2.7 µm  
 Bottom: HALO 160Å ES-C18, 2.7 µm  
 Part Numbers:  
 Top: 92122-604  
 Middle: 92112-606  
 Bottom: 92122-602  
 Mobile Phase A: Water/10 mM difluoroacetic acid  
 Mobile Phase B: ACN/10 mM difluoroacetic acid  
 Gradient: 2-50% B in 60 min  
 Flow Rate: 0.3 mL/min  
 Temperature: 60 °C  
 Detection: 220 nm  
 Injection: 5 µL (0.2 mg/mL)  
 Sample: Trastuzumab Tryptic Digest

Figure 2. This figure demonstrates the utility of the unique selectivity of the 160Å Phenyl-Hexyl Peptide phase. The initial separation using HALO 160Å ES-C18 shows inadequate resolution of peaks five and six. The same separation was attempted on a HALO 160Å ES-CN column, which improved the resolution of peaks five and six, but resulted in coelution of peaks three and four. On the contrary, the HALO 160Å Phenyl-Hexyl column showed excellent resolution for all peaks.

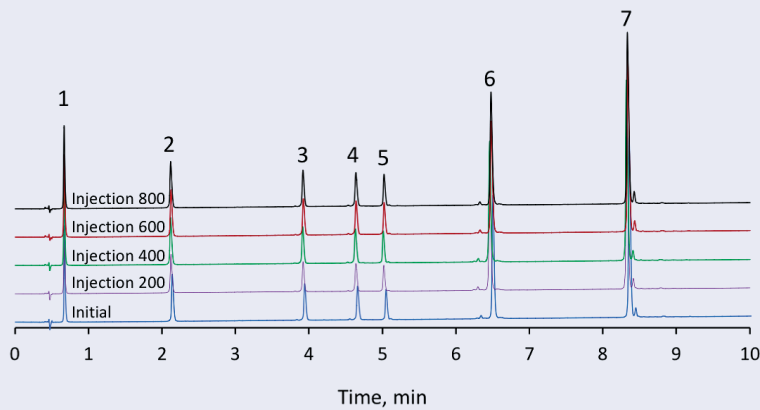


#### TEST CONDITIONS:

Column: 2.1 x 150 mm  
 Top: HALO 160Å ES-C18, 2.7 µm  
 Middle: HALO 160Å ES-CN, 2.7 µm  
 Bottom: HALO 160Å Phenyl-Hexyl, 2.7 µm  
 Part Numbers:  
 Top: 92122-702  
 Middle: 92122-704  
 Bottom: 92112-706  
 Mobile Phase A: 0.1% formic acid in water/10 mM ammonium formate  
 Mobile Phase B: 50/50 n-propanol/water + 0.1% formic acid + 10 mM ammonium formate (pH: 3.45)  
 Gradient: 10-60% B in 15 min  
 Flow Rate: 0.4 mL/min  
 Temperature: 60 °C  
 Detection: 220 nm  
 Injection: 2 µL  
 Sample: (1) tyr-tyr-tyr, (2) angiotensin II, (3) angiotensin 1-12, (4) melittin, (5) sauvagine and (6) β-endorphin

## RUGGED STABILITY

Figure 3. The rugged column stability under low pH and high temperature mobile phase conditions while using a sample containing peptides and small proteins is illustrated in Figure 3. HALO 160Å Phenyl-Hexyl columns can be run at a maximum temperature of 60 °C and with the low pH mobile phase conditions that are typically used for tryptic digests and polypeptides.



### TEST CONDITIONS:

Column: 2.1 x 100 mm

HALO 160Å Phenyl-Hexyl, 2.7 µm

Part Number: 92112-606

Mobile Phase A: Water/0.1% TFA

Mobile Phase B: 70/30 ACN/water/0.1% TFA

Gradient: 9-95% B in 10 min

Flow Rate: 0.5 mL/min

Temperature: 60 °C

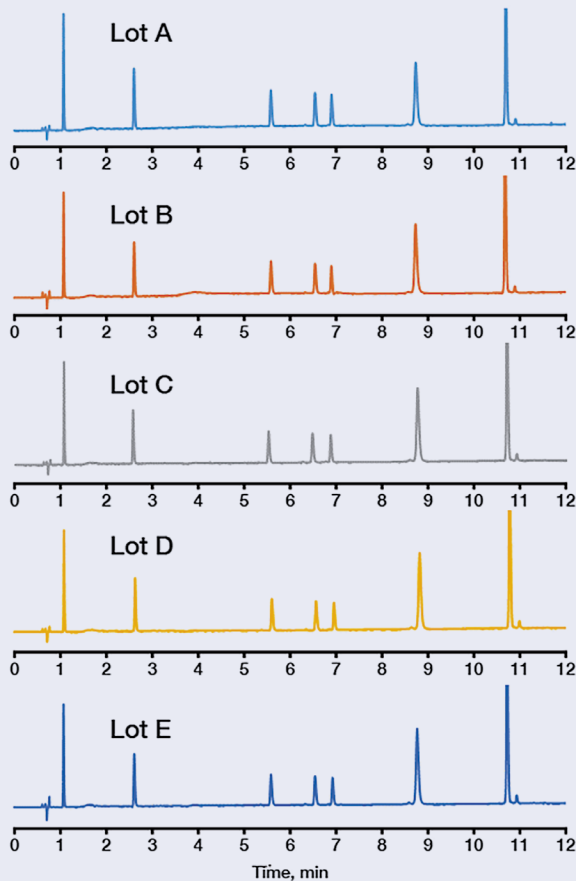
Detection: 220 nm

Injection: 2 µL

Sample: (1) gly-tyr, (2) val-tyr-val, (3) methionine enkephalin, (4) angiotensin II, (5) leucine enkephalin, (6) bovine RNase A and (7) bovine insulin

## LOT-TO-LOT REPRODUCIBILITY

Figure 4. The manufacturing process for HALO 160Å Phenyl-Hexyl bonded phase is tightly controlled to yield repeatable batches. Each lot is quality tested with a mix of peptides and small proteins to ensure lot-to-lot reproducibility. A lot-to-lot comparison is shown below in Figure 4.



### TEST CONDITIONS:

Column: 4.6 x 100 mm

HALO 160Å Phenyl-Hexyl, 2.7 µm

Part Number: 92114-606

Mobile Phase A: 10/90 ACN/water/0.1% TFA

Mobile Phase B: 70/30 ACN/water/0.1% TFA

Gradient: 0-50% B in 15 min

Flow Rate: 1.5 mL/min

Temperature: 30 °C

Detection: 220 nm

Injection: 5 µL

Sample: (1) gly-tyr, (2) val-tyr-val, (3) methionine enkephalin, (4) angiotensin II, (5) leucine enkephalin, (6) bovine RNase A and (7) bovine insulin

**ACT NOW**  
 Contact your local distributor to be among  
 the first to experience these new HALO® columns!

# HALO®

HALO 160Å Phenyl-Hexyl			
Dimension - ID x length (mm)	Part No.	Dimension - ID x length (mm)	Part No.
2.1 x 20	92112-206	3.0 x 100	92113-606
2.1 x 30	92112-306	3.0 x 150	92113-706
2.1 x 50	92112-406	3.0 x 250	92113-906
2.1 x 75	92112-506	4.6 x 20	92114-206
2.1 x 100	92112-606	4.6 x 30	92114-306
2.1 x 150	92112-706	4.6 x 50	92114-406
2.1 x 250	92112-906	4.6 x 75	92114-506
3.0 x 20	92113-206	4.6 x 100	92114-606
3.0 x 30	92113-306	4.6 x 150	92114-706
3.0 x 50	92113-406	4.6 x 250	92114-906
3.0 x 75	92113-506		

HALO 160Å Phenyl-Hexyl Guard Columns, 3/Pack	
Dimension - ID x length (mm)	Part No.
2.1 x 5	92112-106
3.0 x 5	92113-106
4.6 x 5	92114-106
Guard Column Holder (1)	94900-001



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