

160Å ES-C18

160Å PHENYL-HEXYL

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

Enhanced Selectivity With the HALO<sup>®</sup> 160 Å Phenyl-Hexyl Column

**HALO**<sup>®</sup>

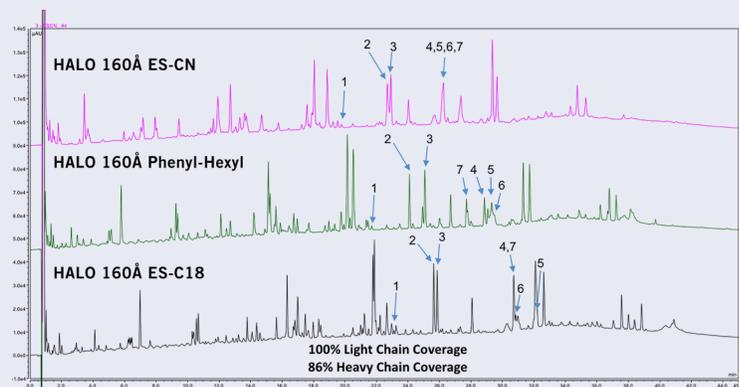
 advancedmaterialstechnology

## INTRODUCING HALO® 160 Å PHENYL-HEXYL

HALO® 160 Å Phenyl-Hexyl columns are specifically designed to offer alternate selectivity to HALO 160 Å ES-C18 and HALO 160 Å 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 160 Å pores enables high resolution at elevated flow rates. Additionally, HALO 160 Å 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 160 Å Phenyl-Hexyl enables different resolutions for tryptic digest fragments compared to HALO 160 Å ES-C18 and HALO 160 Å ES-CN.



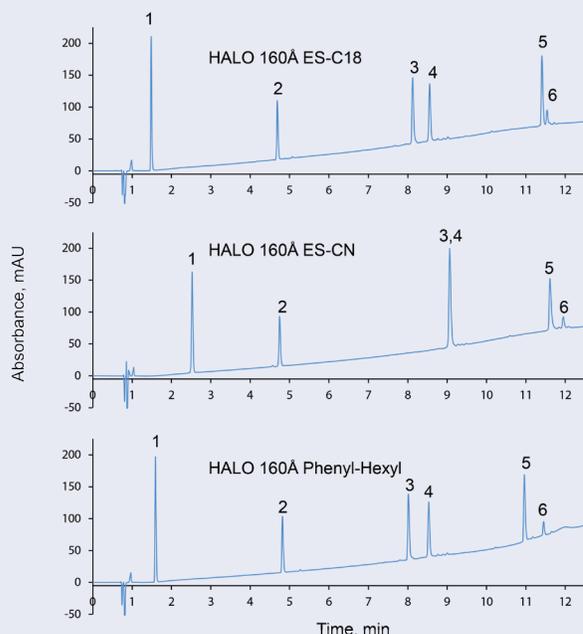
#### PEAK IDENTITIES

- |                           |                                   |
|---------------------------|-----------------------------------|
| 1. FTISADTSKNTAYLQMNSLR   | 5. SGTASWcLLNNFYPR                |
| 2. LScAASGFNIKDTYIHWWR    | 6. ScDKTHTcPPcPAPELLGGPSVFLFPPKPK |
| 3. GFYPSDIAVEWESNGQPENNYK | 7. WSVLTVLHQDWLNGKEYK             |
| 4. LLIYSASFLYSGVPSR       |                                   |

#### TEST CONDITIONS:

**Column:** 2.1 x 100 mm  
**Top:** HALO 160 Å ES-CN, 2.7 µm  
**Middle:** HALO 160 Å Phenyl-Hexyl, 2.7 µm  
**Bottom:** HALO 160 Å 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 160 Å Phenyl-Hexyl Peptide phase. The initial separation using HALO 160 Å ES-C18 shows inadequate resolution of peaks five and six. The same separation was attempted on a HALO 160 Å 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 160 Å Phenyl-Hexyl column showed excellent resolution for all peaks.

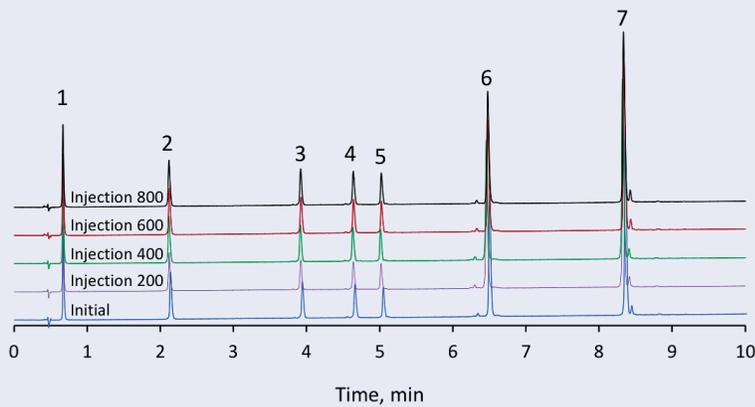


#### TEST CONDITIONS:

**Columns:** 2.1 x 150 mm  
**Top:** HALO 160 Å ES-C18, 2.7 µm  
**Middle:** HALO 160 Å ES-CN, 2.7 µm  
**Bottom:** HALO 160 Å 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 160 Å 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.

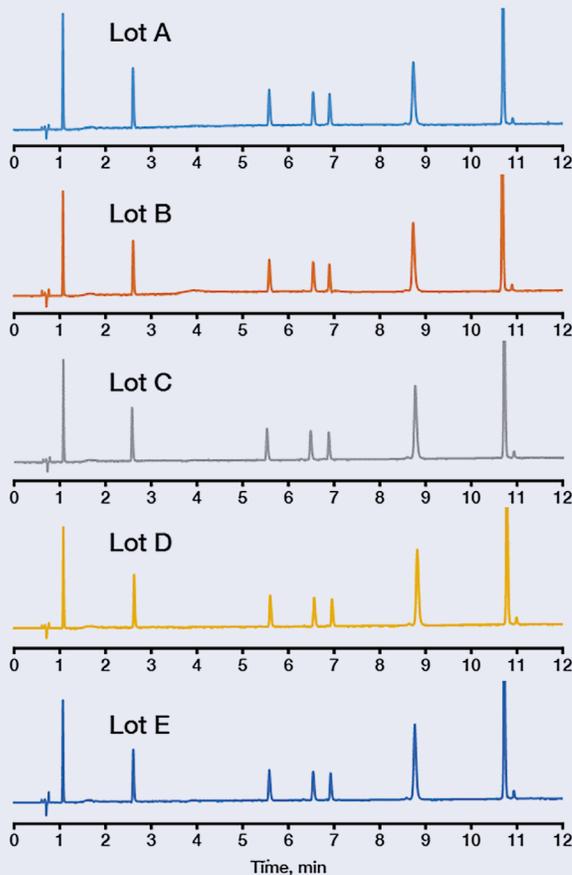


### TEST CONDITIONS:

**Column:** 2.1 x 100 mm  
HALO 160 Å 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 160 Å 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 160 Å 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

# HALO<sup>®</sup>

## BIOCLASS

HALO 160 Å 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 160 Å 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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