

HALO[®]

PCS C18

PEPTIDE

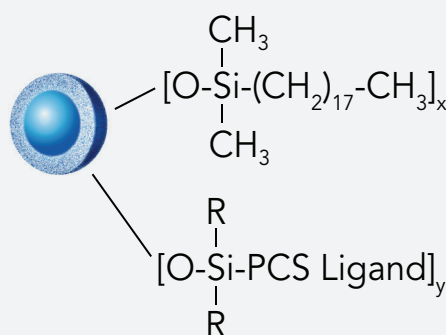
POSITIVE CHARGED TECHNOLOGY
for BASIC COMPOUNDS



HALO[®] PCS (Positive Charged Surface)

POSITIVELY EXCEPTIONAL RESULTS FOR BASIC COMPOUNDS

Built upon proven Fused-Core[®] technology for speed and efficiency, the HALO[®] PCS column products are positively charged surface chemistries designed to deliver improved peak shapes for basic compounds observed with standard C18 chemistries. Ideal for use with low ionic strength mobile phases, HALO[®] PCS maintains peak symmetry at higher loading capacities and provides alternate selectivities from other C18 bonded phases. Available in 160 Å pore size for peptide analysis. The columns are optimized to deliver performance for reproducible, high efficiency LC and LCMS separations.



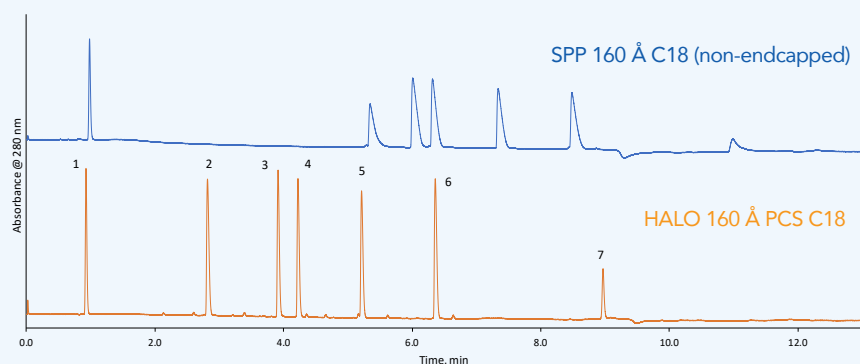
HALO 160 Å PCS C18

FEATURES: PCS C18 for Peptide Separations

- Significantly improved peak widths and symmetry for basic peptides compared to traditional peptide C18 stationary phases
- Designed for performance with formic acid avoiding LCMS signal suppression from TFA
- UHPLC and LCMS compatible
- Alternate L1 selectivity with optimized pore size for peptide separations
- Particle Sizes: 2 and 2.7 μm

THE PCS ADVANTAGE

A synthetic panel of peptides containing basic amino acids is screened on the HALO 160 Å PCS C18 compared to the traditional C18 stationary phase. While using low ionic strength mobile phases such as formic acid the positively charged surface stationary phase shows significantly better peak widths and symmetry for peptides containing basic amino acids when compared to a traditional non-encapped peptide C18 stationary phase.



TEST CONDITIONS:

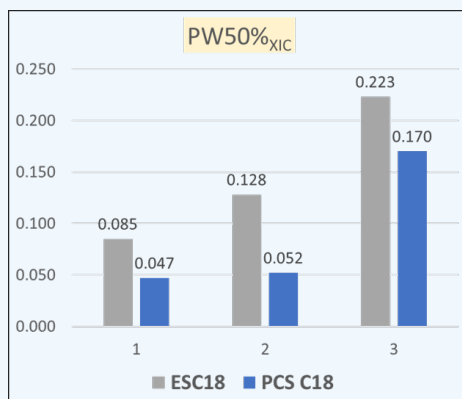
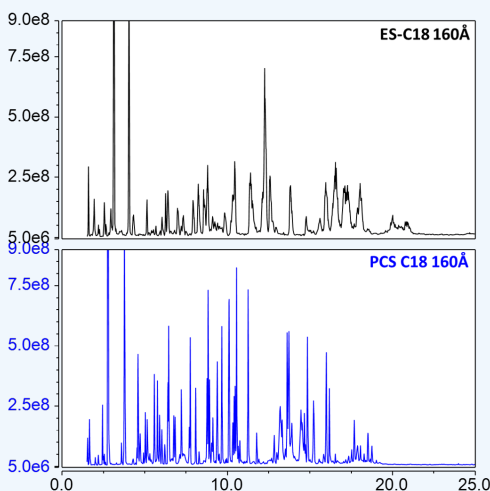
Column: HALO 160 Å PCS C18, 2.7 μm, 2.1 x 100 mm
Part Number: 92812-617
Comparison Column: SPP 160 Å C18, 2.7 μm, 2.1 x 100mm
Mobile Phase A: Water/ 0.1% Formic Acid
Mobile Phase B: Acetonitrile/ 0.1% Formic Acid
Gradient: Time %B
0.0 2
10.0 35
Flow Rate: 0.3 mL/min.
Temperature: 30 °C
Injection Volume: 1.0 μL
Wavelength: PDA, 280 nm
Flow Cell: 1 μL
Data Rate: 100 Hz
Response Time: 0.025 sec.
LC System: Shimadzu Nexera X2

PEAK IDENTITIES:

- | | |
|-------------------------------------|-------------------------------------|
| 1. Uracil | 5. S4Y2 Sequence: Ac-RGVGYLGLGK-NH2 |
| 2. S1Y Sequence: RGAGGLYLGLK-NH2 | 6. S5Y Sequence: Ac-RGVVGLYLGLK-NH2 |
| 3. S2Y Sequence: Ac-RGGGGLYLGLK-NH2 | 7. Insulin Chain B Oxidized |
| 4. S3Y Sequence: Ac-RGAGGLYLGLK-NH2 | |

IMPROVING PEAK WIDTH USING HALO® PCS C18

A separation of Trastuzumab tryptic digest is performed on two HALO® columns, the 160 Å ES-C18 and the 160 Å PCS C18 phases. Significantly narrower peak widths are obtained on the PCS C18 column as shown in the bar graph for three peptides that elute during the beginning, middle, and near the end of the gradient.



TEST CONDITIONS:

Column: HALO 160 Å ES-C18, 2.7 µm, 2.1 x 150 mm

Part Number: 92122-702

Column: HALO 160 Å PCS C18, 2.7 µm, 2.1 x 150 mm

Part Number: 92112-717

Mobile Phase A: Water + 0.1% Formic Acid

Mobile Phase B: Acetonitrile + 0.1% Formic Acid

Gradient:	Time	%B
	0.0	3
	30.0	50
	30.1	95
	33.0	95
	33.1	3
	37.0	3

Flow Rate: 0.4 mL/min.

Pressure: 465 bar

Temperature: 60 °C

Injection Volume: 1 µL

Sample: Trastuzumab Tryptic Digest (1.25 µg/µL)

Sample Solvent: Refer to Digestion Procedure

(halocolumns.com)

LC System: Shimadzu Nexera X2

MS CONDITIONS:

System: QExactive HF

ESI positive polarity

300-2000 m/z

Source voltage: 3.2kV

Sheath Gas: 40

Aux Gas: 20

Aux Gas Temp: 275 °C

Capillary Temp: 320 °C

µscans: 1

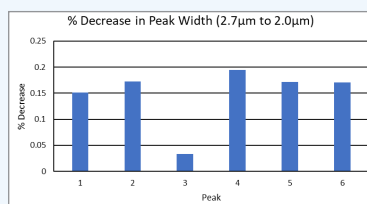
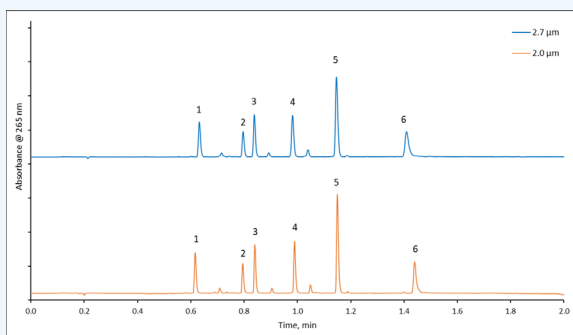
Max Injection Time: 200 msec.

S-Lens RF: 50

#	Tryptic Peptide	XIC	t _R (min)
1	AEDTAVYYC(Carbamidomethyl)SR	667.7877 Z=2	ES-C18: 6.41 PCS C18: 4.60
2	TPEVTC(Carbamidomethyl)VVDVSHEDPEVK	713.6807 Z=3	ES-C18: 12.28 PCS C18: 10.11
3	TVAAPSVFIFPPSDEQLK	973.5171 Z=2	ES-C18: 17.12 PCS C18: 14.47

RAPID PEPTIDE SEPARATION USING 2µm PCS C18

A separation of peptides is performed on two different particle sizes of HALO 160 Å PCS C18 with each column showing excellent peak shape under formic acid conditions. Due to the superficially porous particle technology, flow rates are able to be increased while maintaining column efficiencies allowing for fast, high throughput separations. The decrease in peak width for the 2.0 µm particles is shown in the graph and is accompanied by a corresponding increase in peak height.



TEST CONDITIONS:

Column: HALO 160 Å PCS C18, 2.7 µm, 3.0 x 50 mm

Column: HALO 160 Å PCS C18, 2.0 µm, 3.0 x 50 mm

Mobile Phase A: Water/ 0.1% Formic Acid

Mobile Phase B: Acetonitrile/ 0.1% Formic Acid

Gradient:	Time	% B
	0.0	0
	1.5	35
	2.0	35
	2.1	0
	3.0	0

Flow Rate: 1.5 mL/min.

Pressure: 327 bar - 2.7 µm PCS

651 bar - 2.0 µm PCS

Temperature: 30 °C

Injection Volume: 1.0 µL (0.3 µg/µL)

Wavelength: PDA, 265 nm

Flow Cell: 1 µL

Data Rate: 40 Hz

Response Time: 0.050 sec.

LC System: Shimadzu Nexera X2

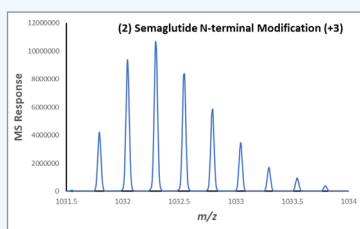
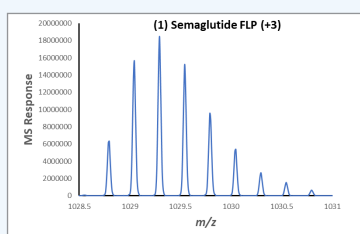
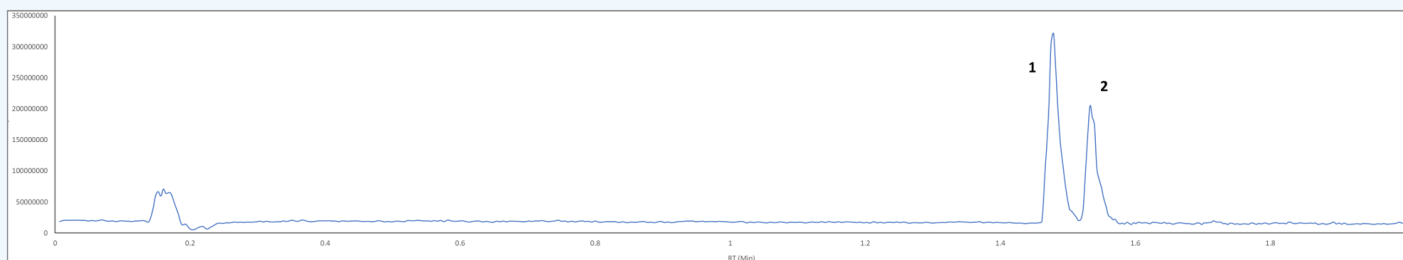
PEAK IDENTITIES:

- | | | | |
|--------|------------------------------|-----------------------------|----------------------------|
| 1. S1Y | Sequence: RGAGGLYL GK-NH2 | 4. S4Y2 | Sequence: Ac-RGVGYL GK-NH2 |
| 2. S2Y | Sequence: Ac-RGGGGLYL GK-NH2 | 5. S5Y | Sequence: Ac-RGVGYL GK-NH |
| 3. S3Y | Sequence: Ac-RGAGGLYL GK-NH2 | 6. Insulin Chain B Oxidized | |

HALO 160 Å PCS C18 PEPTIDE

ULTRAFAST SCREENING FOR SEMAGLUTIDE IMPURITIES USING 2 µm 160 Å PCS C18

The PCS C18 bonded phase contains a positively charged surface ligand in acidic conditions which improves peak shapes in weak ion pairing conditions required for LCMS. Compared to 2.7µm 160 Å PCS C18 in ballistic gradient conditions, peak widths are reduced by approximately 30%, generating peak widths at 50% height to less than 1 second.



Column Type/Sample	Retention Time (min)	50% Peak Width (sec)	Tailing Factor (EP)
2.0 µm PCS Semaglutide FLP	1.476	0.72	1.34
2.0 µm PCS N-terminal Mod	1.533	0.96	1.77
2.7 µm PCS Semaglutide FLP	1.457	1.14	1.4
2.7 µm PCS N-terminal Mod	1.513	1.26	1.69

TEST CONDITIONS:

Column: HALO 160 Å PCS C18, 2.0 µm, 2.1 x 50 mm
HALO 160 Å PCS C18, 2.7 µm, 2.1 x 50 mm

Mobile Phase A: Water + 0.1% Formic Acid

Mobile Phase B: ACN + 0.1% Formic Acid

Gradient: Time	%B
0.0	20
2.0	55
3.0	90
4.0	90

Flow Rate: 0.7 mL/min.

Back Pressure: 2.0µm - 340 bar
2.7µm - 200 bar

Temperature: 60 °C

Injection: 1µL of 20ng Semaglutide modified with 10mM Tris pH 8.0

Sample Solvent: H₂O

LC System: Shimadzu Nexera X2

MS System: Thermo Orbitrap QE-HF

MS CONDITIONS:

Polarity: Positive

Resolution: 60k

AGC Target: 3e6

Max IT: 200ms

Scan Range: 300-2000 m/z

Sheath Gas Flow Rate: 35

Aux Gas Flow Rate: 15

Sweep Gas Flow Rate: 1

Spray Voltage: 4.0kV

Capillary Temp: 375°C

Aux Gas Heater Temp: 350 °C

S-Lens RF level: 60

In-Source CID: 10 eV

TRASTUZUMAB TRYPTIC DIGEST PEAK CAPACITY 2.7µm vs 2µm 160 Å PCS C18

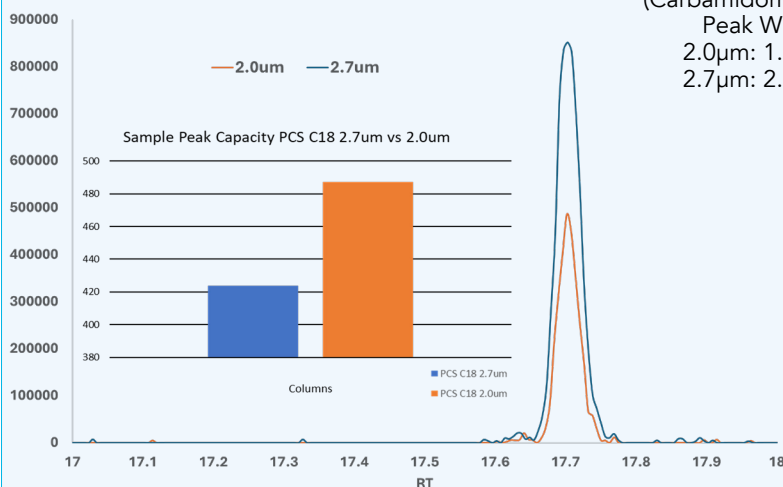
This application compares the performance of two HALO 160 Å PCS C18 columns with different particle sizes, 2 µm and 2.7 µm, using a trastuzumab digest. The 2 µm particle size demonstrated significantly narrower peak widths (~15%), resulting in improved resolution and a substantial increase in peak capacity compared to the 2.7 µm column. The difference in peak capacity between the two columns was 64, highlighting the advantage of smaller particle sizes for complex peptide separations.

PEPTIDE: DYFPEPVTWSNSGALTSGVHTFPAVLQSSGLYSLSVWTPSSSLGTQTYICNVNHPNNTK
(Carbamidomethylation)

Peak Widths:

2.0µm: 1.92 SEC

2.7µm: 2.22 SEC



TEST CONDITIONS:

Column: HALO 160 Å PCS C18, 2.0 µm, 2.1 x 100 mm

Mobile Phase A: Water + 0.1% Formic Acid

Mobile Phase B: Acetonitrile + 0.1% Formic Acid

Gradient: Time	%B
0.0	3
30.0	50
35.0	3

Flow Rate: 0.3 mL/min

Back Pressure: 330 bar

Temperature: 60 °C

Injection: 1µL of 200 ng/mL

Trastuzumab Digest

Sample Solvent: H₂O

LC System: Shimadzu Nexera X2

MS System: Thermo Q-Exactive HF

MS CONDITIONS:

Polarity: Positive

Resolution: 120k

AGC Target: 3e6

Max IT: 200ms

Scan Range: 200-2000 m/z

Sheath Gas Flow Rate: 25

Aux Gas Flow Rate: 10

Sweep Gas Flow Rate: 1

Spray Voltage: 4.5kV

Capillary Temp: 350 °C

Aux Gas Heater Temp: 300 °C

S-Lens RF level: 60



PRODUCT CHARACTERISTICS

ATTRIBUTE	160 Å PCS C18
Ligand	dimethyloctadecylsilane
Particle Size (µm)	2.0, 2.7
Pore Size (Å)	160
USP #	L1
Carbon Load (%)	4.2, 5.0
Surface Area(m²/g)	68, 90
Endcapped (Y/N)	Yes
Low pH Limit/Max T	2/60 °C
High pH Limit/Max T	7/40 °C
100% Aqueous Compatible	Yes

PART NUMBERS

Dimensions: ID x Length (in mm)	160 Å PCS C18 (2µm)	160 Å PCS C18 (2.7µm)
1.5 x 50	9118X-417	9211X-417
1.5 x 100	9118X-617	9211X-617
1.5 x 150	9118X-717	9211X-717
2.1 x 20	91182-217	
2.1 x 30	91182-317	
2.1 x 50	91182-417	92112-417
2.1 x 100	91182-617	92112-617
2.1 x 150	91182-717	92112-717
2.1 x 250	91182-917	
3.0 x 30	91183-317	
3.0 x 50	91183-417	92113-417
3.0 x 100	91183-617	92113-617
3.0 x 150	91183-717	92113-717
3.0 x 250	91183-917	
4.6 x 50		92114-417
4.6 x 100		92114-617
4.6 x 150		92114-717

HALO® GUARD COLUMNS 3 PACK

Dimensions: ID x Length (in mm)	160 Å PCS C18 (2µm)	160 Å PCS C18 (2.7µm)
2.1 x 5	91182-117	92112-117
3.0 x 5	91183-117	92113-117
4.6 x 5		92114-117
Guard Column Holder	94900-001	

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AMT26_PCS_PeptideREV0

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