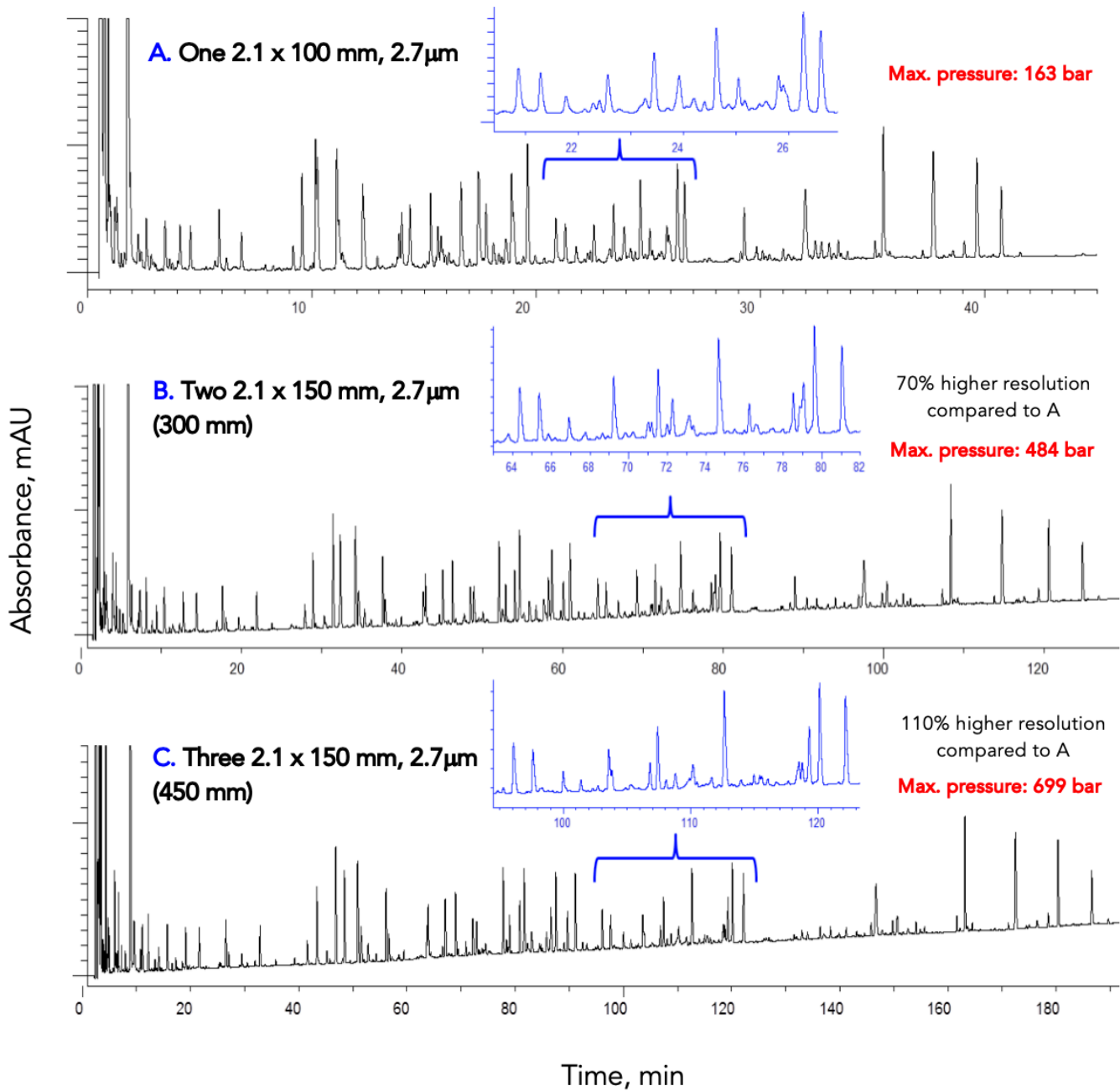




Analysis of Apotransferrin Tryptic Digest on HALO® 160 Å Columns

Application Note 179-PE



**TEST CONDITIONS:****Columns:**

1) HALO 160 Å ES-C18, 2.7 µm, 2.1 x 100 mm

Part Number: 92122-602

2) HALO 160 Å ES-C18, 2.7 µm, 2.1 x 150 mm

Part Number: 92122-702**Mobile Phase:**

A: Water with 0.1% TFA

B: 80/20 acetonitrile/water with 0.1% TFA

Flow Rate: 0.4 mL/min**Temperature:** 60 °C**Detection:** UV 215 nm, PDA**Injection Volume:** 10 µL**Sample Solvent:** Water**Response Time:** 0.05 sec**Data Rate:** 40 Hz**Flow Cell:** 1.0 µL**LC System:** Shimadzu Nexera X2

Gradient A:	Time (min)	% B
	0.0	5
	60	60

Gradient B:	Time (min)	% B
	0.0	5
	180	60

Gradient C:	Time (min)	% B
	0.0	5
	270	60

The chromatograms on the preceding page show a comparison of an apotransferrin tryptic digest sample analyzed on three different lengths of HALO® 160 Å ES-C18 columns: a single 2.1 x 100 mm, two 2.1 x 150 mm columns in series, and three 2.1 x 150 mm columns in series. The insets show examples of the improved performance obtained using longer column lengths along with longer gradient times for demanding samples. Resolution increases of approximately 70% and 110% are achieved by increasing column length by 3-fold and 4.5-fold respectively. Gradient times of 60, 180 and 270 minutes were used for the top, middle and bottom chromatograms, respectively.

Lower pressures afforded by both 2.7 and 5 µm HALO® Peptide particles allow two or more columns to be used in series for additional resolution and peak capacity for challenging peptide mapping analyses. HALO® 160 Å ES-C18 is also available in 2.0 µm particle sizes in 2.1 and 3 mm IDs up to 150 mm length for additional options in run time and peak capacity.

