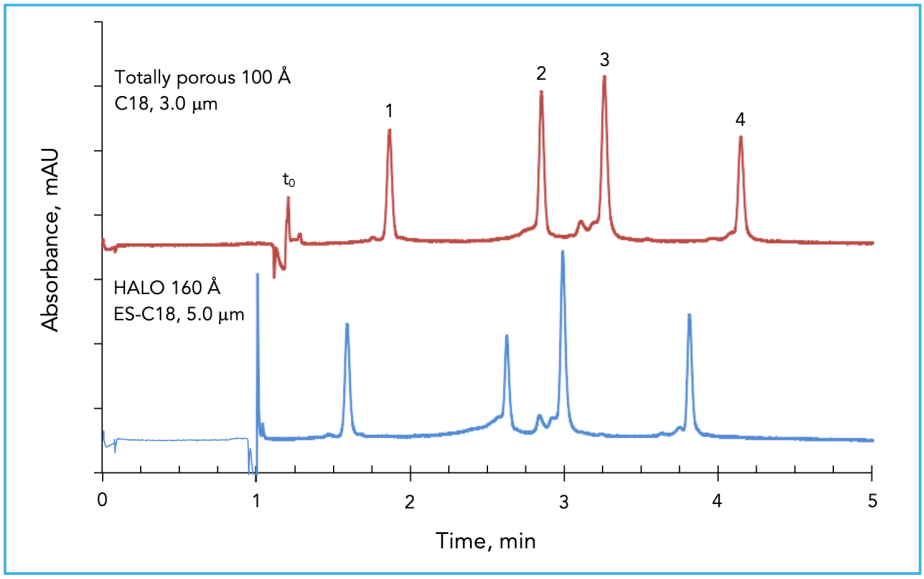
HALO®

BIOPHARMACEUTICALS

Separation of Four Small Proteins on HALO® 160 Å ES-C18, 5 µm vs. Totally Porous C18, 3.0 µm

Application Note 104-PR



PEAK IDENTITIES:

1. Ribonuclease A (13.7 KDa)
2. Cytochrome c (12.4 KDa)
3. Lysozyme (14.3 KDa)
4. α-Lactalbumin (14.2 KDa)

These chromatograms show the separation of four low MW proteins on HALO 160 Å ES-C18, 5 µm column vs. a totally porous C18, 3.0 µm column. The separations are similar with the benefit of the HALO® 5 µm column having lower back pressure and similar resolution. The HALO® 5 µm ES-C18 phase is made with sterically hindered silanes during manufacture, enhancing the stability-even at temperatures up to 90 °C. The stability of the totally porous C18 column was not evaluated.

TEST CONDITIONS:

Columns:
1) HALO 160 Å ES-C18, 5 µm, 4.6 x 150 mm
Part Number: 95124-702
2) 100 Å totally porous C18, 3.0 µm, 4.6 x 150 mm
Mobile Phase: 72/28 - A/B (start)
A: Water with 0.1% trifluoroacetic acid
B: Acetonitrile with 0.1% trifluoroacetic acid
Gradient: 28% B to 55% B in 5 min
Flow Rate: 1.5 mL/min
Pressure: 95 bar (HALO®)
170 bar (competitor)
Temperature: 60 °C
Detection: UV 280 nm, PDA
Injection Volume: 15 µL
Sample Solvent: Mobile phase A
Response Time: 0.1 sec
Flow Cell: 2.0 µL micro cell
LC System: Agilent 1200 SL