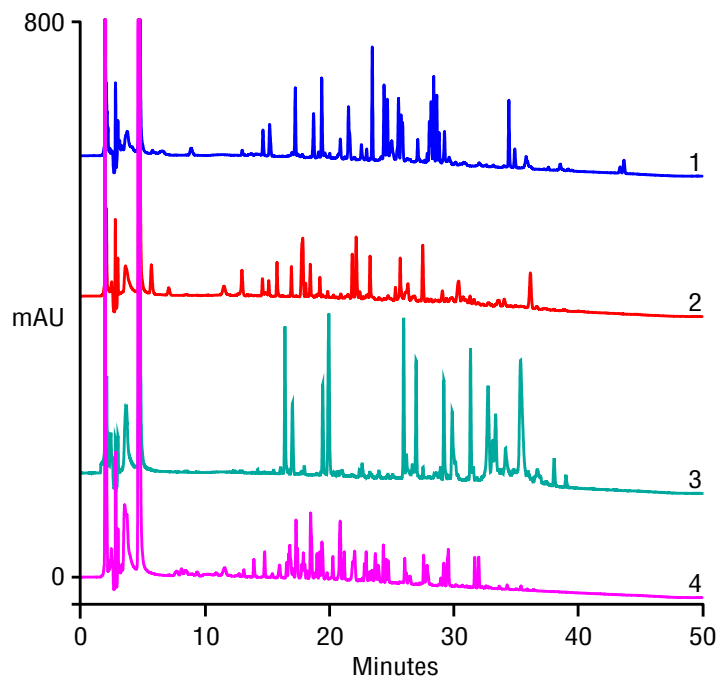


Four Peptide Maps on Acclaim PolarAdvantage II (PA2)



Column: Acclaim® PA2, 3 µm
 Dimensions: 3.0 x 250 mm
 System: UltiMate® 3000 RSLC
 Mobile Phases: A: 300 µL TFA + 45 mg Na₄P₂O₇•10H₂O in 1.00 L water
 B: 300 µL TFA + 546 g CH₃CN + 300 g water

Gradient Times:
 min -10 0 7 45 50
 %A 93 93 93 30 30
 %B 7 7 7 70 70

Flow Rate: 0.60 mL/min
 Injection: 20 µL
 Temperature: 30 °C
 Detection: Diode array, UV 210 (shown), 280 nm

Samples:
 1. Ovalbumin
 2. Fetuin
 3. β-Casein
 4. Bovine serum albumin

Preparation:
 0.6 mg protein
 Denaturation with 8 M urea
 Reduction with tributyl phosphine
 Alkylation with iodoacetamide
 Digestion with trypsin (Promega Corp.)
 Final volume 0.4 mL

26204

Mapping tryptic protein digests can produce highly complex chromatograms where every peak counts. The Acclaim 3 µm 3.0 × 250 mm columns yield superior peak capacity and resolution. Using techniques developed for Rapid Separation LC (RSLC) columns, these columns are physically rugged and rated to 800 bar. The PolarAdvantage II chemistry exhibits excellent chemical stability in acidic conditions, and has complementary selectivity compared to C18. Some peptides, especially phosphopeptides, are sensitive to metal contamination. A small amount of pyrophosphate (0.1 mM) in the mobile phase suppresses metal effects without affecting UV detection.