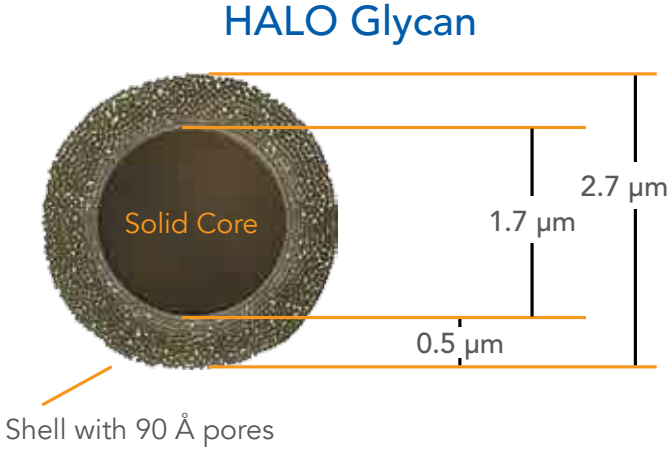
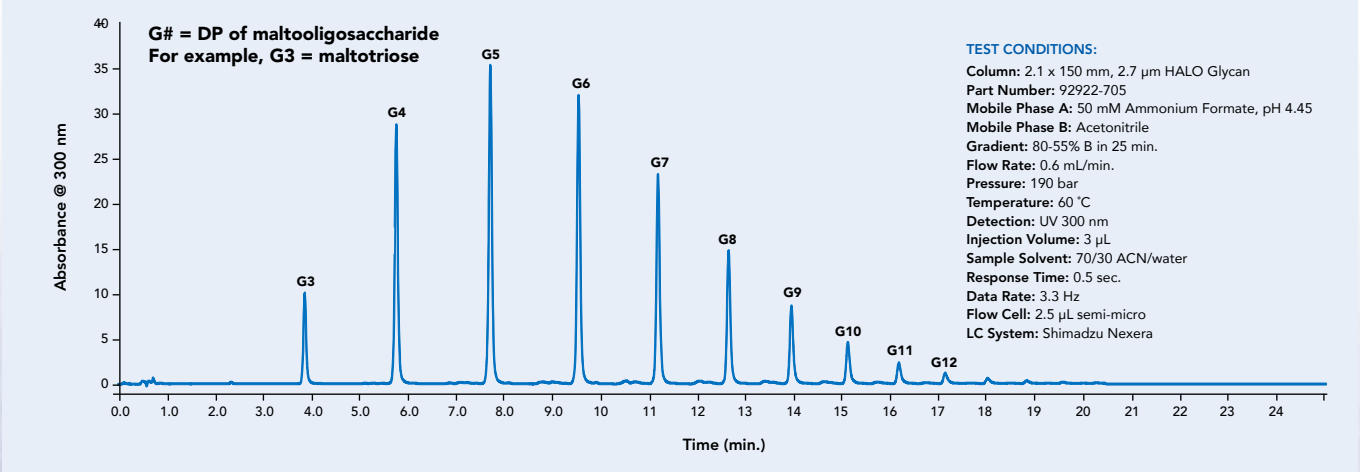
HALO GLYCAN

- 90 Ångstrom pore size
- Incorporates a highly polar ligand that contains 5 hydroxyl groups tethered to 2.7 µm Fused-Core silica particles via novel, proprietary linkage chemistry (Table F)
- Ideal for hydrophilic interaction liquid chromatography (HILIC) separations of oligosaccharides, and particularly, of released and labeled glycans from glycoproteins and proteoglycans
- Mobile phases typically consist of acetonitrile and aqueous ammonium formate buffer (50 mM, pH 4.4) used to form a gradient of increasing water content during elution
- Each lot of HALO Glycan material is tested for quality assurance (Figure KK) by separation of a procainamide-reducing-end-labeled glycan ladder of oligosaccharides having 2–25 glucose units (GU).
  - Peaks for oligosaccharides composed of 5 and 10 GU must meet tight specifications for retention and peak width before lot is approved for glycan analysis
- 2 µm inlet frit
- Pressure limit, 600 bar/9000 psi



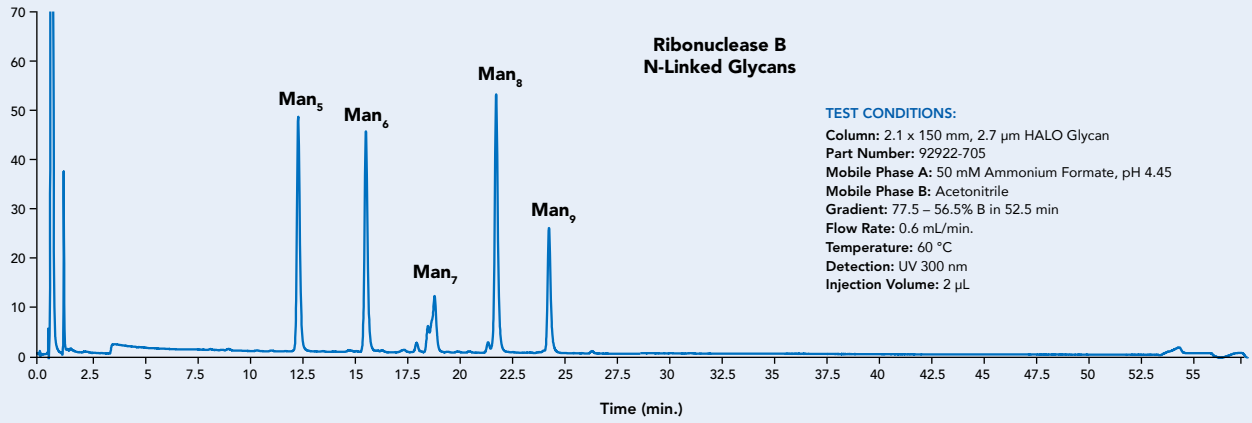
QA ANALYSIS OF HALO GLYCAN

Figure KK. Example QA Chromatogram for HALO Glycan column. Each HALO Glycan packing lot is tested using this glycan ladder mixture to assess and ensure lot-to-lot reproducibility.



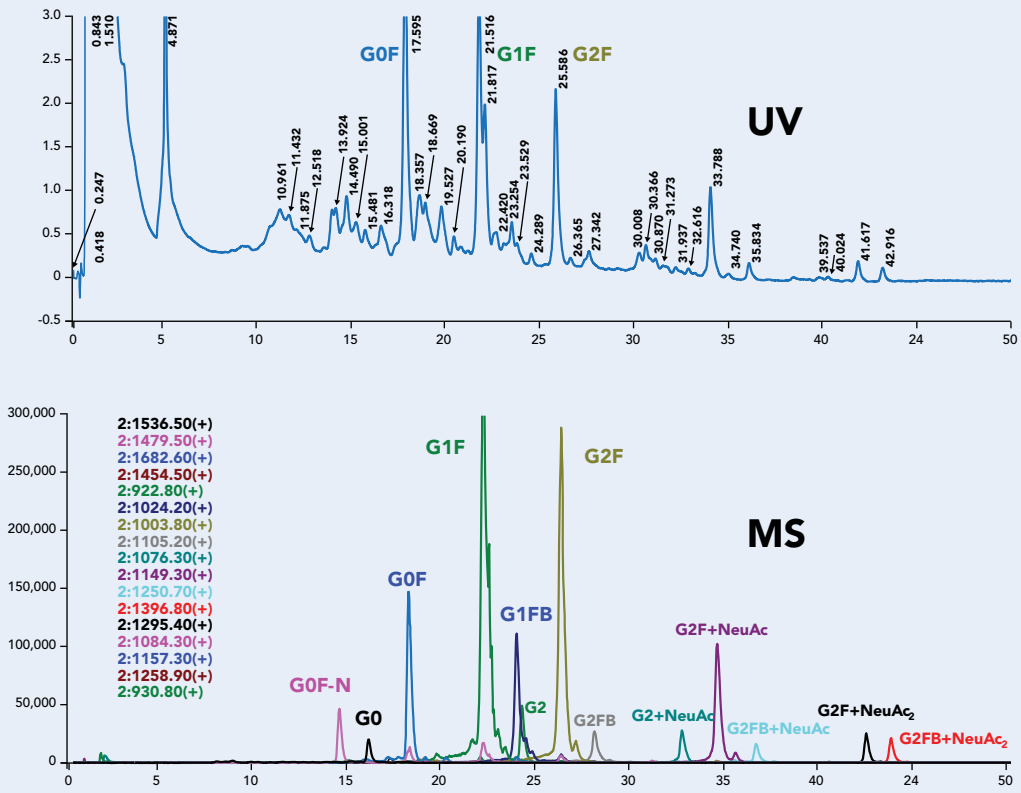
SEPARATION OF N-LINKED GLYCANS FROM RIBONUCLEASE B

Figure LL. Gradient HILIC-MS separation of N-linked glycans, which had been released using PNGase from ribonuclease B, using the HALO Glycan column.



SEPARATION OF N-LINKED GLYCANS FROM HUMAN IgG

Figure MM. Released- and procainamide-labeled glycans from human IgG were separated using a 2.1 x 150 mm HALO Glycan column and detected using UV and selected-ion-monitoring MS detection.



TEST CONDITIONS:
Column: 2.1 x 150 mm, HALO 2.7 µm Glycan
Part Number: 92922-705
Mobile Phase A: 50 mM Ammonium Formate, pH 4.4
Mobile Phase B: Acetonitrile
Gradient: 77.5 – 60% B in 60 min
Flow Rate: 0.5 mL/min.
Temperature: 60 °C
Detection: UV 300 nm
Injection Volume: 4 µL
LC System: Shimadzu Nexera
MS: Shimadzu LCMS 2020 (single quadrupole)
ESI: +4.7 kV
Scan range: 500-2000 m/z
Scan rate: 2 pps

See page 35 for full list of HALO Glycan part numbers.