

Chromatography Solutions

Avantor® ACE® Excel® Glycan

UHPLC/HPLC columns for reproducible glycan analysis



Avantor® ACE®

GLYCAN ANALYSIS

- Analysis of the glycosylation pattern of biopharmaceutical compounds is essential for their complete characterisation
- Monoclonal antibodies (mAbs) are one of the largest growing areas of biotherapeutic interest and are heavily glycosylated molecules with each mAb containing at least two glycosylation sites as shown in Figure 1 below
- Changes to the glycosylation of a biopharmaceutical can affect the efficacy and safety of the drug, so it is extremely important that it is thoroughly monitored
- Due to the highly polar nature of released glycans, a hydrophilic stationary phase is ideal for their separation
- Labelling with a fluorophore such as 2-aminobenzamide (2-AB) allows the released glycans to be analysed using fluorescence detection

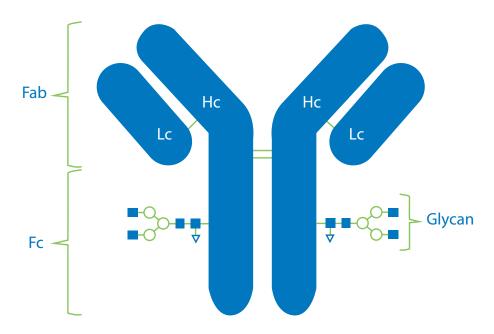


Figure 1: Schematic showing glycosylation regions of a monoclonal antibody.

Phase	Endcapped				Carbon load (%)	Recommended pH range
ACE® Excel® Glycan	No	1.7, 3	100	300	7.0	2.0 - 7.0

To extend column lifetime ACE UHPLC guard cartridges (Cat. No. EXL-1P16-GD2U / Cat. No. EXL-1P16-GD4U) are recommended.

For the guard system, guard cartridge holder (H0011) is also required.

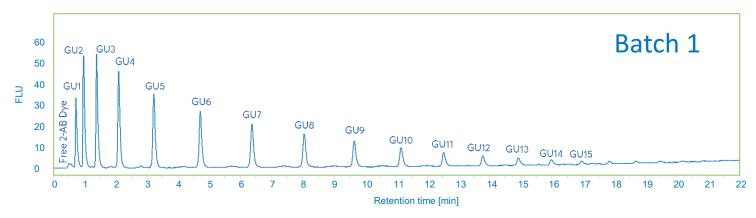
For HPLC column connections up to 6000 psi, PEEK™ fingertight fittings (Cat. No. ACE-CC10) are recommended - for further details please contact **chromsupport@avantorsciences.com**

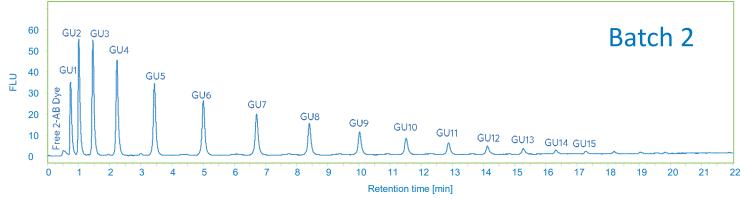
For UHPLC column connections up to 25000 psi, reuseable fittings (Cat. No. EXL-CC10) are recommended - for further details please contact **chromsupport@avantorsciences.com**

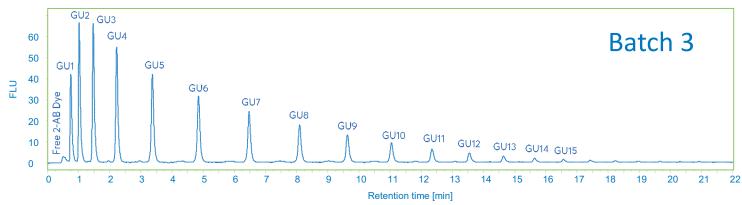
GUARANTEED REPRODUCIBILITY

- The ACE® Excel® Glycan column provides excellent batch-to-batch reproducibility as shown below
- The dextran ladder standard consists of a mixture of glucose homopolymers of between 1 to 15 monomer units, and is a useful tool in identifying more complex glycoforms based upon correlated retention times

BATCH-TO-BATCH REPRODUCIBILITY







Sample: 2-AB labelled Dextran Ladder standard

Column: ACE Excel 1.7 Glycan, 100x2.1 mm **Cat. No.:** EXL-1716-1002

Mobile phase: A = 100 mM ammonium formate (pH 4.5) and B = MeCN

Gradient: 75 – 52.5% B in 24 min **Flow rate:** 0.5 ml/min Temperature: 55 °C

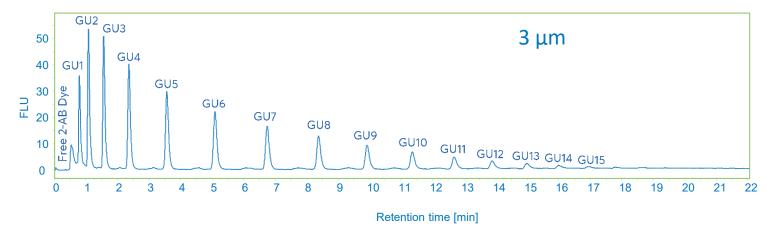
Injection: 1 μ l Detection: FLD, λ_{ex} = 260 nm, λ_{em} = 430 nm

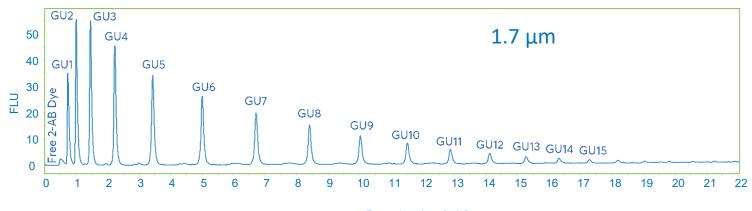
AVANTOR® ACE® EXCEL® GLYCAN

GUARANTEED SCALABILITY

The ACE® Excel® Glycan column provides exceptionally reproducible scalability between the 1.7 µm and 3 µm particle sizes, as demonstrated below

PARTICLE SIZE COMPARISON





Retention time [min]

Sample: 2-AB labelled Dextran Ladder standard
Columns: ACE Excel 1.7 Glycan, 100x2.1 mm; ACE Excel 3 Glycan, 100x2.1 mm
Cat. No.: EXL-1716-1002 and EXL-1716-1002
Mobile phase: A = 100 mM ammonium formate (pH 4.5) and B = MeCN
Gradient: 75 - 52.5% B in 24 min

Flow rate: 0.5 ml/min

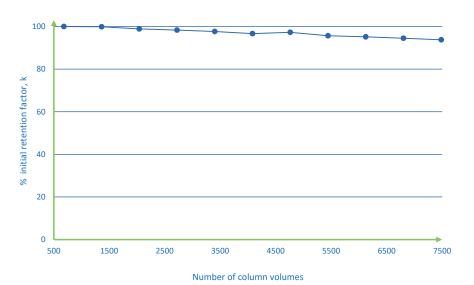
Temperature: 55 °C Injection: 1 µl

Detection: FLD, $\lambda_{ex} = 260 \text{ nm}$, $\lambda_{em} = 430 \text{ nm}$

EXCELLENT COLUMN STABILITY

- The ACE® Excel® Glycan column shows good stability for glycan analysis over several thousand column volumes as seen in the graph below

COLUMN STABILITY UNDER GLYCAN ANALYSIS CONDITIONS



Sample: 2-AB labelled Dextran Ladder standard Column: ACE Excel 1.7 Glycan, 50x2.1 mm

Cat. No.: EXL-1716-0502

Mobile phase: A = 100 mM ammonium formate (pH 4.5) and B = MeCN Gradient: 75 – 60% B in 8 min

Flow rate: 0.5 ml/min Temperature: 55 °C Injection: 0.5 µl

Detection: FLD, λ ex = 260 nm, λ _{em} = 430 nm

AVANTOR® ACE® EXCEL® GLYCAN UHPLC/HPLC ORDERING INFORMATION

Avantor® ACE® Excel® Glycan columns

Column dimensions	1.7 µm	3 μm
50 x 2.1 mm	EXL-1716-0502	EXL-1116-0502
100 x 2.1 mm	EXL-1716-1002	EXL-1116-1002
150 x 2.1 mm	-	EXL-1116-1502
50 x 4.6 mm	-	EXL-1116-0546
100 x 4.6 mm	-	EXL-1116-1046
150 x 4.6 mm	-	EXL-1116-1546

Avantor® ACE® Excel® Glycan guard cartridges

Description	Cat. No.
For analytical columns (2.1 mm ID)	EXL-1P16-GD2U
For analytical columns (4.6 mm ID)	EXL-1P16-GD4U

Related products

Description	Cat. No.
UHPLC guard holder	H0011
ACE Excel UHPLC column connector	EXL-CC10