PROzyme
A part of Agilent

CERTIFICATE OF ANALYSIS

PRODUCT NAME:	AdvanceBio 2-AB CHO mAb N-Linked Glycan Library
PRODUCT CODE:	GKSB-020
LOT NUMBER:	DP18E0103a
PACK SIZE:	200 pmol (qualitative chromatographic standard for N-glycan identification)
FORM:	Dry solid
STORAGE:	Store at -20°C in the dark before and after reconstitution
EXPIRATION:	June 2024, may be used for 1 year after reconstitution
STRUCTURE:	The 2-AB CHO mAb Library contains N-glycans whose reducing termini are derivatized with the fluorescent dye, 2-AB (2-aminobenzamide).

3832 Bay Center Place
Hayward, CA 94545-3619

TOLL FREE (800) 457-9444
PHONE (510) 638-6900
FAX (510) 638-6919

info@prozyme.com
www.prozyme.com

Quality Control:

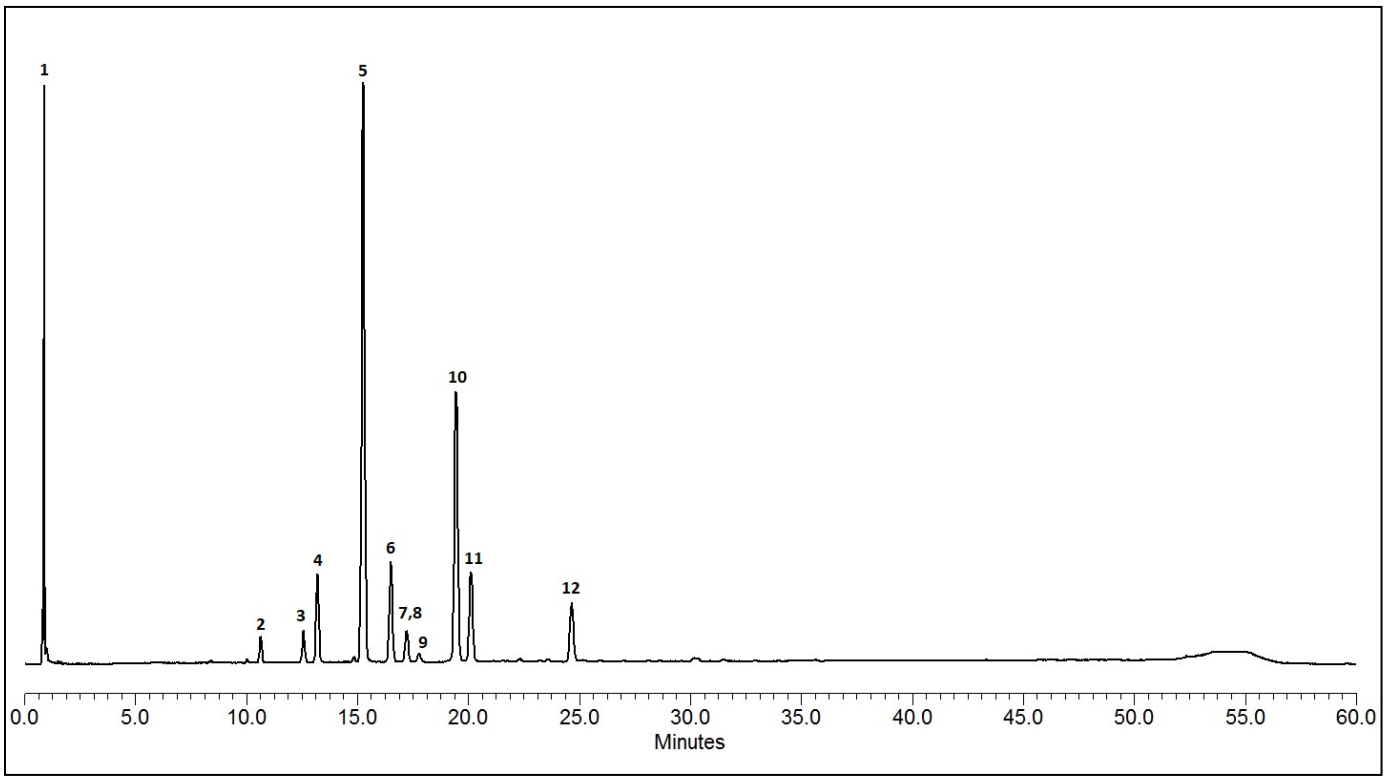
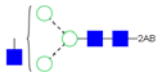
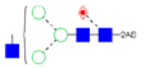
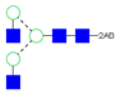
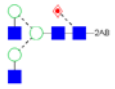
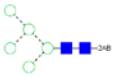
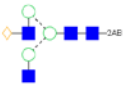
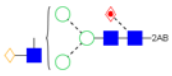
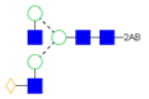
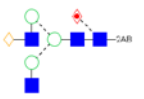
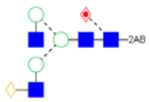
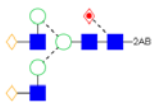


Figure 1 - **UPLC® Results**: 1 µl (aqueous) of the 2-AB-labeled glycan library was injected on a Waters ACQUITY UPLC H Class System utilizing a 60-minute method under the conditions below (see Table 1 for peak ID; the number of peaks observed depends on the running conditions employed):

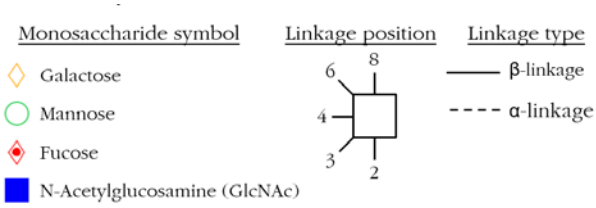
Time (min)	Flow (ml/min)	%ACN	%Buffer A
0.00	0.4	80.0	20.0
2.00	0.4	80.0	20.0
2.50	0.4	75.0	25.0
50.0	0.4	62.0	38.0
52.0	0.4	40.0	60.0
53.5	0.4	40.0	60.0
55.0	0.4	80.0	20.0
60.0	0.4	80.0	20.0

Column: Waters ACQUITY UPLC BEH Glycan Column (1.7 µm, 2.1 x 150 mm)
Buffer A: 100 m*M* ammonium formate, pH 4.4
ACN: Acetonitrile
Flow rate: As stated in table, in ml/min
Temperature: 60° C
Max Pressure: 15,000 psi
Fluorescence Detection: λ_{ex} = 360 nm, λ_{em} = 428 nm

Table 1 - Peak Identification of 2-AB CHO mAb Library

Peak No.	Glycan Identification	ProZyme	Common	Oxford (New)	Structure[4,5]
1	Free Dye (2-AB)				
2	Asialo-, agalacto-, biantennary, -1 N-Acetylglucosamine	NGA2-N	G0-N	A1	
3	Asialo-, agalacto-, biantennary with core fucose, -1 N-Acetylglucosamine	NGA2F-N	G0F-N	F(6)A1	
4	Asialo-, agalacto-, biantennary	NGA2	G0	A2	
5	Asialo-, agalacto-, biantennary with core fucose	NGA2F	G0F	F(6)A2	
6	Oligomannose 5	Man-5	Man5	M5	
7	Asialo-, mono-galactosylated, biantennary	NA2G1[6]	G1[6]	A2[6]G(4)1	
8	Asialo-, mono-galactosylated, biantennary with core fucose, -1 N-Acetylglucosamine	NA2G1F-N	G1F-N	F(6)A1G(4)1	
9	Asialo-, mono-galactosylated, biantennary	NA2G1[3]	G1[3]	A2[3]G(4)1	
10	Asialo-, mono-galactosylated, biantennary with core fucose	NA2G1F[6]	G1F[6]	F(6)A2[6]G(4)1	
11	Asialo-, mono-galactosylated, biantennary with core fucose	NA2G1F[3]	G1F[3]	F(6)A2[3]G(4)1	
12	Asialo-, galactosylated biantennary with core fucose	NA2F	G2F	F(6)A2G(4)2	

Structure Key[1,2]:



Structural Analysis: The purity and structural integrity of the glycan library was assessed by UPLC[3] (as described above) and MALDI-TOF[4,5] or LC-MS. Agreement was found between the results from mass spectrometry and UPLC.

Application:

- Qualitative standard for various analytical procedures
- As a migration standard for liquid chromatography

Handling & Reconstitution: The labeled oligo-saccharide library is shipped as a dried solid. Use ultra-pure water or an aqueous buffer to dissolve the materials (see Directions for Use for suggested volumes).

Allow the unopened vial to reach ambient temperature and tap on a solid surface to ensure that most of the material is at the bottom of the vial. Gently remove the cap, add the desired volume of ultra-pure water or aqueous buffer, re-cap and mix thoroughly to redissolve all the material. For maximal recovery, ensure that the cap lining is also rinsed. Centrifuge the reconstituted vial briefly before use.

Make sure that any glassware, plasticware, solvents or reagents used are free of glycosidases and carbohydrate contaminants.

Minimize exposure to elevated temperatures or extremes of pH.

Store the reconstituted glycan library at -20° C. Allow the vial to equilibrate to ambient temperature before use.

Directions For Use: The amount of 2-AB-labeled glycan standard injected on a UPLC column is typically 1 µl (6 – 9 pmol of total glycan). The number of injections obtained from each vial depends on the reconstitution and subsequent dilution (if any) of the library.

For our Quality Control testing, one vial was dissolved in 30 µl of water and 1 µl injected on the ACQUITY column. For larger injection volumes or other LC systems we recommend further dilution as necessary for compatibility with your mobile phase. For suggested methods see Rapid UPLC Methods for Screening Labeled N-Glycans at:

www.prozyme.com/protocols/

REFERENCES

1. Ceroni A, Maass K, Geyer H, Geyer R, Dell A, Haslam SM. GlycoWorkbench: a tool for the computer-assisted annotation of mass spectra of glycans. J Proteome Res. 2008 Apr; 7(4): 1650-9.

2. Harvey DJ, Merry AH, Royle L, Campbell MP, Dwek RA, Rudd PM. Proposal for a standard system for drawing structural diagrams of N– and O-linked carbohydrates and related compounds. Proteomics 2009 Aug; 9(15): 3796-801.

3. Ahn J, Bones J, Yu YQ, Rudd PM, Gilar M. Separation of 2-aminobenzamide labeled glycans using hydrophilic interaction chromatography columns packed with 1.7 microm sorbent. J Chromatogr B Analyt Technol Biomed Life Sci. 2010 Feb 1; 878(3-4): 403-8.

4. James DC, Jenkins N. Analysis of N-glycans by matrix-assisted laser desorption/ionization mass spectrometry; in A laboratory guide to glycoconjugate analysis. BioMethods (P. Jackson and J. T. Gallagher, ed) 1997; 9: 91-112.

5. Papac DI, Wong A, Jones AJS. Analysis of acidic oligosaccharides by matrix-assisted laser desorption/ionization time of flight mass spectrometry. Anal Chem 1996 Sep 15; 68(18): 3215-3223.

Digitally signed by Shiva Pourkaveh
Date: 2019.08.06 17:08:48 -07'00'

Authorized Signature