CERTIFICATE OF ANALYSIS

PRODUCT NAME:	GLYKO® 2-AB-(BIANTENNARY & HIGH MANNOSE PARTITIONED LIBRARY)
PRODUCT CODE:	GKSB-520
LOT NUMBER:	P14J1401
PACK SIZE:	200 pmol of each library (qualitative chromatographic standard for N-glycan identification) 1 each WS0311 2-AB-(Fucosyl Biantennary Library) 1 each WS0312 2-AB-(Afucosyl Biantennary Library) 1 each WS0313 2-AB-(High Mannose Library)
FORM:	Dry solid
STORAGE:	Store in the dark at -20°C before and after reconstitution
EXPIRATION:	January 2024, may be used for 1 year after reconstitution
STRUCTURE:	The 2-AB-(Biantennary & High Mannose Partitioned Library) consists of 3 blended libraries of N-linked glycans whose reducing termini are derivatized with the fluorescent dye, 2-AB (2-aminobenzamide). The libraries were partitioned to minimize overlap of peaks to facilitate glycan peak identification.

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Quality Control:

UPLC Running Conditions: 3 pmol (1 µl) of each 2-AB labeled glycan library was injected on a UPLC BEH Glycan Column, 1.7 µm 2.1 x 100 mm from Waters utilizing a 10-minute method under the conditions below:

Time (min)	Flow (ml/min)	% ACN	% Buffer
0	1.0	75	25
8.0	1.0	60	40
8.1	0.5	40	60
8.5	0.5	40	60
8.6	1.0	40	60
8.8	1.0	75	25
10.0	1.0	75	25

ACN: acetonitrile

Buffer: 100 m*M* ammonium formate, pH 4.4

Temperature: 60°C

Max. Pressure: 15,000 psi

Fluorescence Detection: λ_{ex} = 330 nm λ_{em} = 420 nm

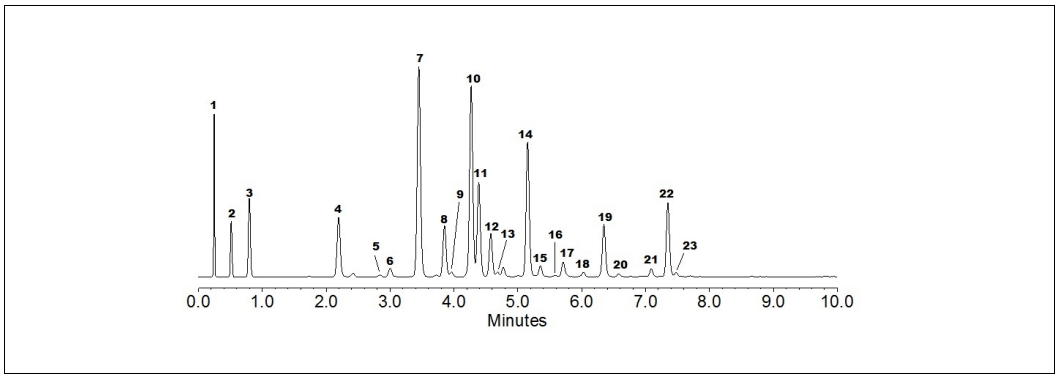

Figure 1. ACQUITY UPLC® Results of WS0311 - See Table 1 for peak ID.

Table 1: Peak Identification of WS0311 2-AB-(Fucosyl Biantennary Library)

Peak Number	Glycan Identification
1	Free Dye (2-AB)
2	6-α-fucosyl chitobiose (N2F)
3	4'-β-mannosylchitobiose, core-α(1-6)-substituted with fucose (MNNF)
4	Conserved trimannosyl core, substituted with fucose (M3N2F)
5	Conserved trimannosyl core, substituted with fucose + N-Acetylglucosamine (M3N2F + GlcNAc)
6	Asialo-, agalacto- biantennary (NGA2/G0)
7	Asialo-, agalacto- biantennary, core-substituted with fucose (NGA2F/G0F)
8	Asialo-, agalacto- biantennary, core-substituted with fucose and with bisecting N-Acetylglucosamine (NGA2FB/G0FB)
9	Asialo, mono-galactosylated biantennary (NA2G1/G1)
10 + 11	Asialo, mono-galactosylated biantennary, core substituted with fucose (NA2G1F/G1F)
12 + 13	Asialo, mono-galactosylated biantennary, core substituted with fucose and with bisecting N-Acetylglucosamine (NA2G1FB/G1FB)
14	Asialo-, galactosylated biantennary, core-substituted with fucose (NA2F/G2F)
15	Asialo-, galactosylated biantennary, core-substituted with fucose and with bisecting N-Acetylglucosamine (NA2FB/G2FB)
16 + 17	Mono-sialylated, mono-galactosylated, biantennary core-substituted with fucose (G1FS1)
18	Mono-sialylated, galactosylated biantennary (A1)
19	Mono-sialylated, galactosylated biantennary, core-substituted with fucose (A1F)
20	Mono-sialylated, galactosylated biantennary, core-substituted with fucose and with bisecting N-Acetylglucosamine (A1FB)
21	Di-sialylated, galactosylated biantennary (A2)
22	Di-sialylated, galactosylated biantennary, core-substituted with fucose (A2F)
23	Di-sialylated, galactosylated biantennary, core-substituted with fucose and with bisecting N-Acetylglucosamine (A2FB)

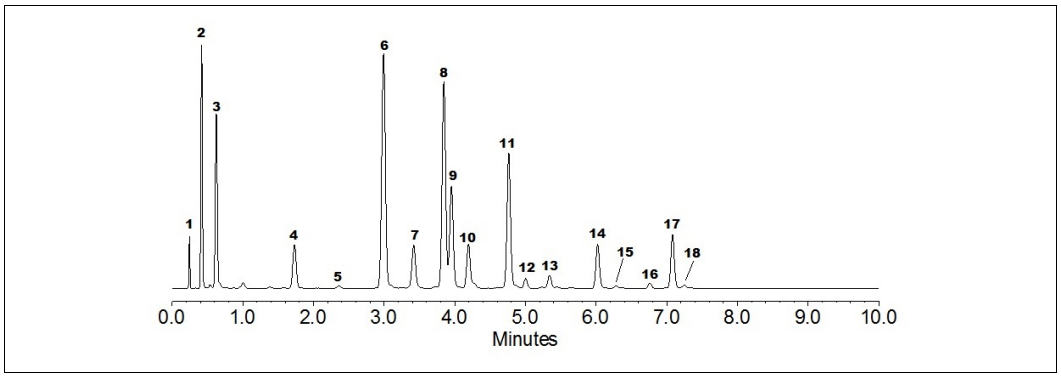

Figure 2. ACQUITY UPLC® Results of WS0312 - See Table 2 for peak ID.

Table 2: Peak Identification of WS0312 2-AB-(Afucosyl Biantennary Library)

Peak Number	Glycan Identification-
1	Free Dye (2-AB)
2	Chitobiose (N2/NN)
3	4'-β-mannosylchitobiose (MNN)
4	Conserved trimannosyl core (M3N2/Man-3)
5	Conserved trimannosyl core (M3N2/Man-3) + N-Acetylglucosamine (GlcNAc)
6	Asialo-, agalacto- biantennary (NGA2/G0)
7	Asialo-, agalacto- biantennary with bisecting N-Acetylglucosamine (NGA2B/G0B)
8 + 9	Asialo, mono-galactosylated biantennary (NA2G1/G1)
10	Asialo, mono-galactosylated biantennary,with bisecting N-Acetylglucosamine (NA2G1B/G1B)
11	Asialo-, galactosylated biantennary (NA2/G2)
12	Asialo-, galactosylated biantennary with bisecting N-Acetylglucosamine (NA2B/G2B)
13	Mono-sialylated, mono-galactosylated biantennary (G1S1)
14	Mono-sialylated, galactosylated biantennary (A1)
15	Mono-sialylated, galactosylated biantennary with bisecting N-Acetylglucosamine (A1B)
16 + 17	Di-sialylated, galactosylated biantennary (A2)
18	Di-sialylated, galactosylated biantennary with bisecting N-Acetylglucosamine (A2B)

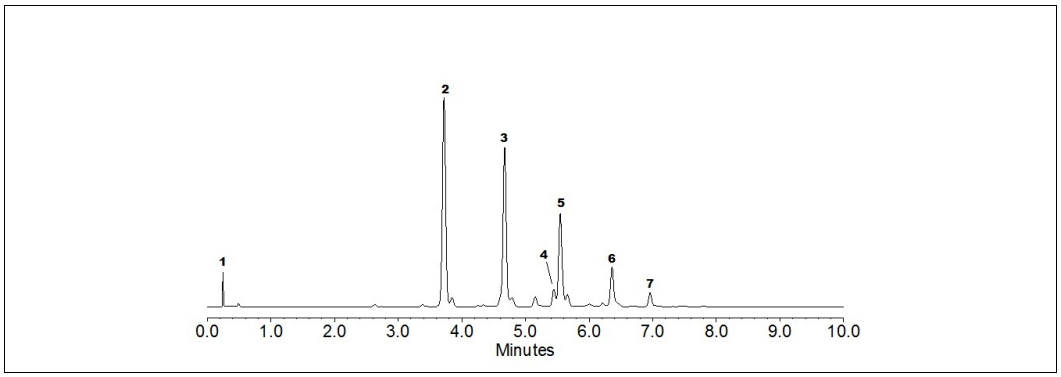

Figure 3. ACQUITY UPLC® Results of WS0313 - See Table 3 for peak ID.

Table 3: Peak Identification of WS0313 2-AB-(High Mannose Library)

Peak Number	Glycan Identification
1	Free Dye (2-AB)
2	Oligomannose 5 (Man-5)
3	Oligomannose 6 (Man-6)
4 + 5	Oligomannose 7 (Man-7)
6	Oligomannose 8 (Man-8)
7	Oligomannose 9 (Man-9)

Structural Analysis: The purity and structural integrity of the glycan libraries were assessed by a combination of methods including UPLC (GU values) and LC/MS.

Application: As a peak reference standard for liquid chromatography.

Handling: The labeled oligosaccharide is shipped as a dried solid. 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 water or buffer, re-cap and mix thoroughly to redissolve all the oligosaccharide. For maximal recovery, ensure that the cap lining is also rinsed and 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.

Reconstitution: Use HPLC-grade water or an aqueous buffer to dissolve the glycan to the desired concentration as described in the directions for use below. Store the reconstituted glycan at -20°C. Avoid multiple freeze/thaw cycles.

Directions For Use: The amount of 2-AB-labeled library standard injected on the ACQUITY UPLC BEH Glycan Column is typically 3 - 6 pmol of total glycan. 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/

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