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CERTIFICATE OF ANALYSIS

PRODUCT NAME:	GLYKO® APTS HUMAN IgG N-LINKED GLYCAN LIBRARY
PRODUCT CODE:	GKSP-005
LOT NUMBER:	P14A2001c
PACK SIZE:	120 pmol (qualitative capillary electrophoresis standard for glycan identification)
FORM:	Dry solid
STORAGE:	Store at -20°C in the dark before and after reconstitution
EXPIRATION:	November 2022, may be used for 3 months after reconstitution
STRUCTURE:	The APTS-(Human IgG N-Linked Glycan Library) consists of complex biantennary oligosaccharides consistent with N-glycans on normal human IgGs[1,2,3] with the reducing termini derivatized with the fluorescent dye, APTS (8-Aminopyrene-1,3,6-trisulfonic acid trisodium salt).

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Quality Control:

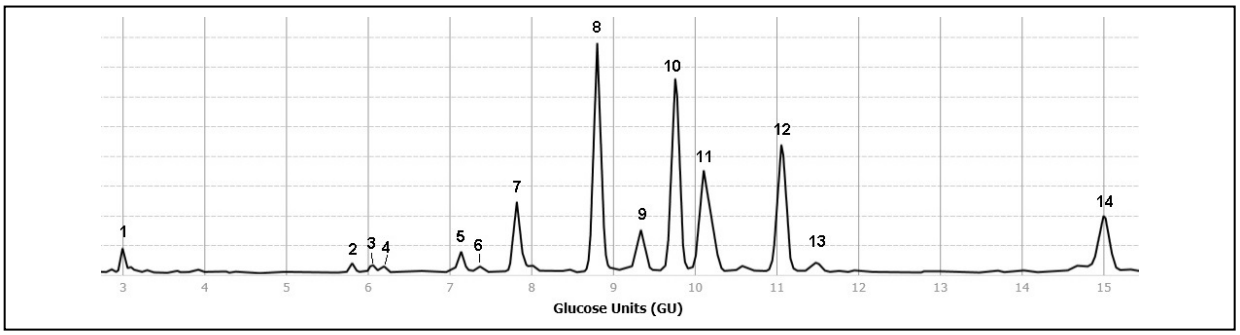
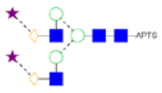
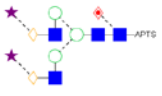
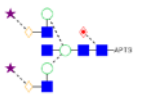
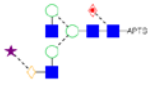


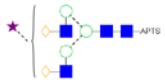
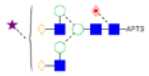
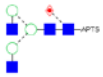
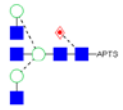
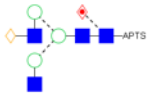
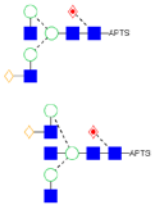
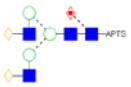
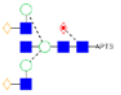
Figure 1 – Gly-Q™ Results: The APTS labeled standard is injected onto a Gly-Q CE Instrument (GQ2100) under the conditions and method below (for a representative electropherogram of this library run on an AB Sciex PA800 *plus* Pharmaceutical Analysis System, see last page):

METHOD:

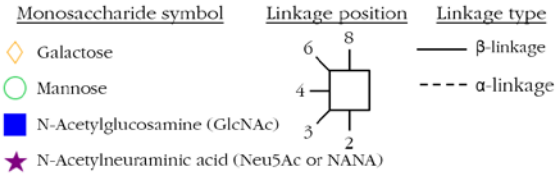
Action	High Voltage	Duration, seconds	Position	Sampling Interval, Seconds
High Voltage Purge	4.00	10.00	Wash	0.04
Pause		2.00	Clean	
Reagent Block Injection	2.00	2.00	MA02	0.04
Pause		2.00	Clean	
Well Plate Injection	2.00	2.00	Sample	0.04
Pause		2.00	Clean	
Separation & Detection	10.00	120.00	Separation	0.04

Table 1 –Peak Identification of APTS-(Human IgG N-Linked Glycan Library). Gly-*Q* Migration Standards (GKSQ-500) were co-injected with the library. Peaks are listed in order of appearance on the electropherogram (left to right).

Peak Number	Glycan Identification	ProZyme	Common	Oxford (New)	Structure [5,6]
1	GKSQ-500 Lower Migration Standard (LMS), DP3				
2	Di-α(2-6)-sialylated, galactosylated biantennary	A2	G2S2	A2G(4)2S(6)2	
3	Di-α(2-6)-sialylated, galactosylated biantennary with core fucose	A2F	G2FS2	F(6)A2G(4)2S(6)2	
4	Di-α(2-6)-sialylated, galactosylated biantennary, with core fucose and with bisecting GlcNAc	A2FB	G2FS2B	F(6)A2BG(4)2S(6)2	
5	Mono-α(2-6)-sialylated, galactosylated biantennary with core fucose	NA2G1FS1	G1FS1	F(6)A2[3]G(4)1S(6)1	

Peak Number	Glycan Identification	ProZyme	Common	Oxford (New)	Structure [5,6]
6	Mono-α(2-6)-sialylated, galactosylated biantennary	A1	G2S1	A2G(4)2S(6)1	
7	Mono-α(2-6)-sialylated, galactosylated biantennary, with core fucose	A1F	G2FS1	F(6)A2G(4)2S(6)1	
8	Asialo-, agalacto-biantennary with core fucose	NGA2F	G0F	F(6)A2	
9	Asialo-, agalacto-biantennary with core fucose and with bisecting GlcNAc	NGA2FB	G0FB	F(6)A2B	
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 + Asialo-, mono-galactosylated biantennary, with core fucose and with bisecting GlcNAc	NA2G1F[3] + NA2G1FB[6]	G1F[3] + G1FB[6]	F(6)A2[3]G(4)1 + F(6)A2[6]BG(4)1	
12	Asialo-, galactosylated biantennary, with core fucose	NA2F	G2F	F(6)A2G(4)2	
13	Asialo-, galactosylated biantennary, with core fucose and with bisecting GlcNAc	NA2FB	G2FB	F(6)A2BG(4)2	
14	GKSQ-500 Upper Migration Standard (UMS), DP15				

Structure Key[4,5]:



Structural Analysis: The purity and structural integrity of the glycan library was assessed by CE analysis. The identity of the unlabeled glycan library is confirmed by MALDI-TOF[6,7] mass spectrometry or LC-MS. Agreement was found between the CE results and published GU values[8].

Application:

- As a peak reference standard for capillary electrophoresis

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. Avoid multiple freeze/thaw cycles.

Directions For Use: The amount of labeled glycan standard used with the Gly-Q Instrument is typically 50 µl. For our Quality Control testing, the dried glycan was dissolved in 100 µl of water and an aliquot was then further diluted 1:100. 100 µl of the diluted glycan was transferred to a well of a 96-well PCR plate and replicate runs were processed. Typically, ~30 injections are obtained from a 50 µl sample. Signal decrease can occur over repeated injections from the same aliquot. Conditions may vary for other CE systems, for assistance, please contact technical service at:

info@prozyme.com

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Authorized Signature

PA800 Electropherogram (representative)

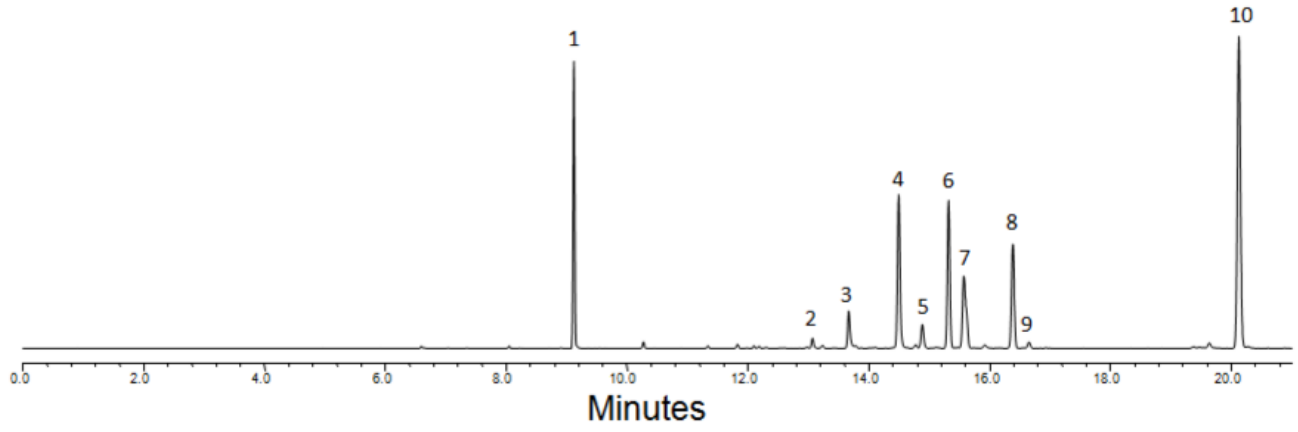

Figure 2. The APTS-labeled glycan library was injected on an AB Sciex PA800 *plus* Pharmaceutical Analysis System under the conditions listed below:

Capillary	N-CHO Capillary, length to window 50 cm; total length 60 cm (AB Sciex P/N 477601)
Electroylyte	Carbohydrate Separation Buffer (AB Sciex P/N 477623)
Pressure	0.5 psi
Time	10 seconds
Capillary Temp.	20°C
Field Strength	600 V/cm
Fluorescence Detection	λ_{ex} = 488 nm
	λ_{ex} = 520 nm

Table 2: Peak Identification of APTS-Labeled Human IgG N-Linked N-Glycan Library on PA800

Peak Number on PA800	Glycan Identity
1	GKSP-500 Bracketing Standard, DP2 (maltose)
2	Mono-sialylated, mono-galactosylated, biantennary core-substituted with fucose [G1FS1[3]]
3	Mono-sialylated, galactosylated biantennary, core-substituted with fucose [A1F (2,6)] and Asialo-, agalacto- biantennary [NGA2/G0]
4	Asialo-, agalacto- biantennary, core-substituted with fucose [NGA2F/G0F]
5	Asialo-, agalacto- biantennary, core-substituted with fucose and with bisecting N-Acetylglucosamine [NGA2FB/G0FB]
6	Asialo, mono-galactosylated biantennary, core substituted with fucose [NA2G1F [6]/G1F [6]]
7	Asialo, mono-galactosylated biantennary, core substituted with fucose [NA2G1F [3]/G1F [3] and Asialo, mono-galactosylated biantennary, core substituted with fucose and with bisecting N-Acetylglucosamine [NA2G1FB [6]/G1FB [6]]
8	Asialo-, galactosylated biantennary, core-substituted with fucose [NA2F/G2F]
9	Asialo-, galactosylated biantennary, core-substituted with fucose and with bisecting N-Acetylglucosamine [NA2FB/G2FB]
10	GKSP-500 Bracketing Standard, DP15 (maltopentadecaose)