



Certificate of Analysis

IgG Heavy (¹³C) Permethylated Glycan Library

Cat. #: CPM-C13-IGG-01

Batch #: B351-01

Size: approx 20 MS runs

Description: A mixture of fucosylated, bi-antennary glycan standards with variable sialylation released from human IgG antibody glycoprotein and permethylated.

Source: The glycans in this product are released from an IgG standard that is purified from human serum. IgG exists in a variety of glycoforms containing bi-antennary oligosaccharides with variable sialylation.

Form: Dry. Lyophilised powder.

Storage: Refrigerate (-20°C) both before and after dissolving. This product is stable for at least 5 years as supplied.

Shipping: The product is shipped at ambient temperature.

Handling: Once dissolved, avoid repeated thawing and refreezing, storage over 3 h at room temperature or above, exposure to light and long term exposure to acid as these will cause glycan desialylation.

Safety: This product is non-hazardous and has been purified from natural sources certified to be free of all hazardous material including pathogenic biological agents.

For research use only. Not for human or drug use

Data: CPM-C13-IGG-01_B351-01-1-pos-850-0001.A13[c] 10 May 2013 9:12 Cal: RD-B35A-01-pos-850- 10 May 2013 9:07
 Shimadzu Biotech Axima Resonance 2.9.0.20090423: Mode positive, Mid 850+, Power: 93
 103 mV[sum= 7757 mV] Profiles 1-75 Unsmoothed

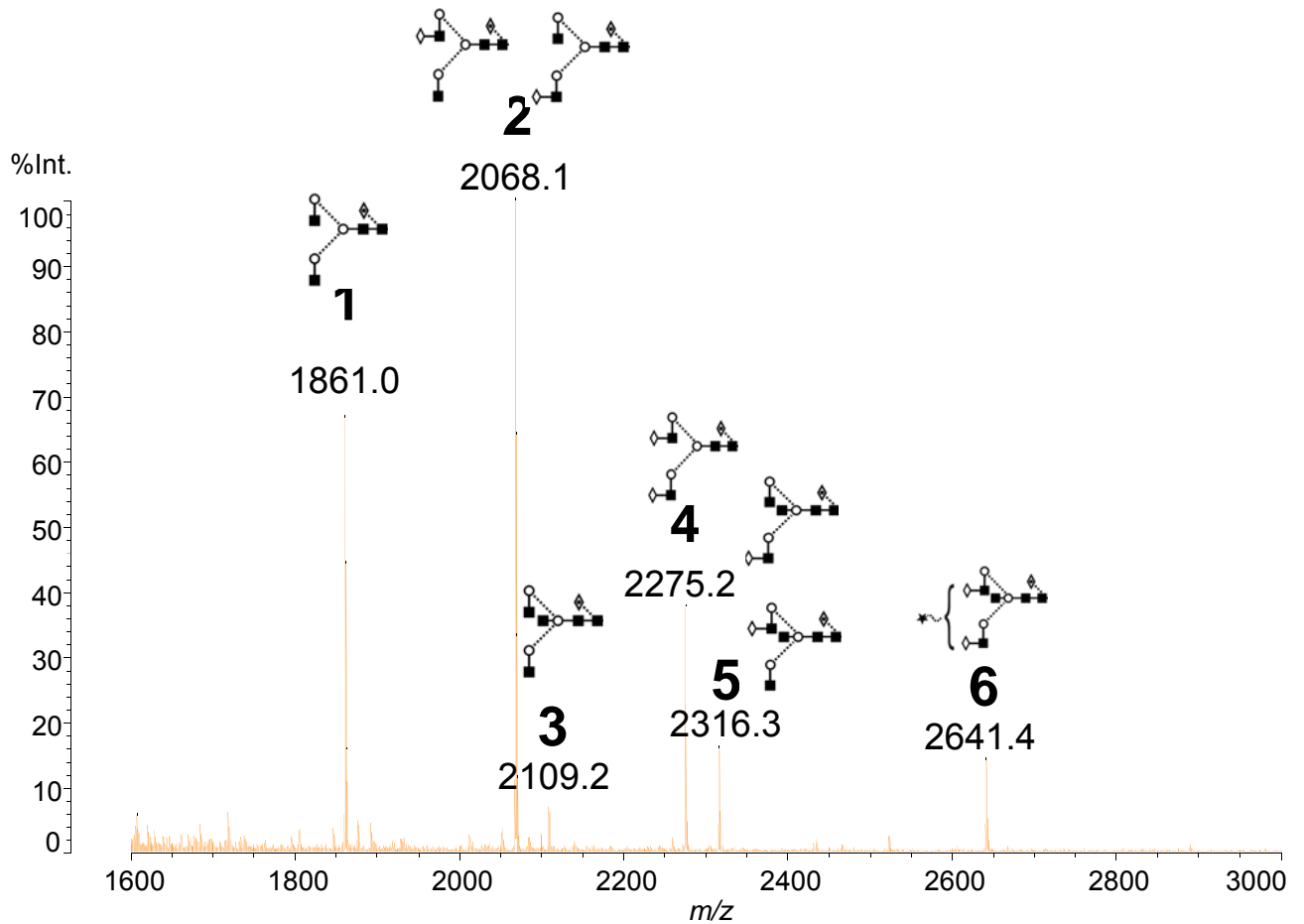


Figure 1: Mass spectrum of heavy (^{13}C)permethylated IgG N-Glycans standard (Cat. # CPM-C13-IGG-01, Batch # B351-01) released from Human IgG antibody by N-Mode hydrazinolysis. Analysis performed on Shimadzu Biotech Resonance MALDI-Ion Trap with DHB matrix. Table 1 shows peak assignments.

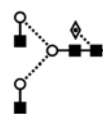
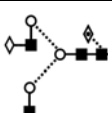
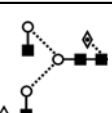
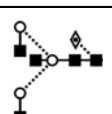
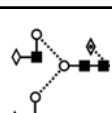

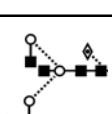
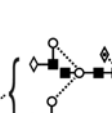
| PeakID | Full Name | Short Name | Structure | ¹³ CPermethylated m/z [M+Na ⁺] | |
|--------|-----------------|------------|---|--|--------|
| | | | | Calculated | Found |
| 1 | F(6)A2 | FA2 |  | 1860.5 | 1861.0 |
| 2 | F(6)A2[6]G(4)1 | FA2G1 |  | 2068.1 | 2068.1 |
| | F(6)A2[3]G(4)1 | |  | | |
| 3 | F(6)A2B | FA2B |  | 2109.2 | 2109.2 |
| 4 | F(6)A2G(4)2 | FA2G2 |  | 2275.2 | 2275.2 |
| 5 | F(6)A2[6]BG(4)1 | FA2BG1 |  | 2316.1 | 2316.3 |
| | F(6)A2[3]BG(4)1 | |  | | |
| 6 | F(6)A2G(4)2S1 | FA2G2S1 |  | 2640.7 | 2641.4 |

Table 1: Structures and names of each peak from the MALDI spectra of permethylated IgG N-Glycans standard (Cat. #: CPM-C13-IGG-01, Batch # B351-01)

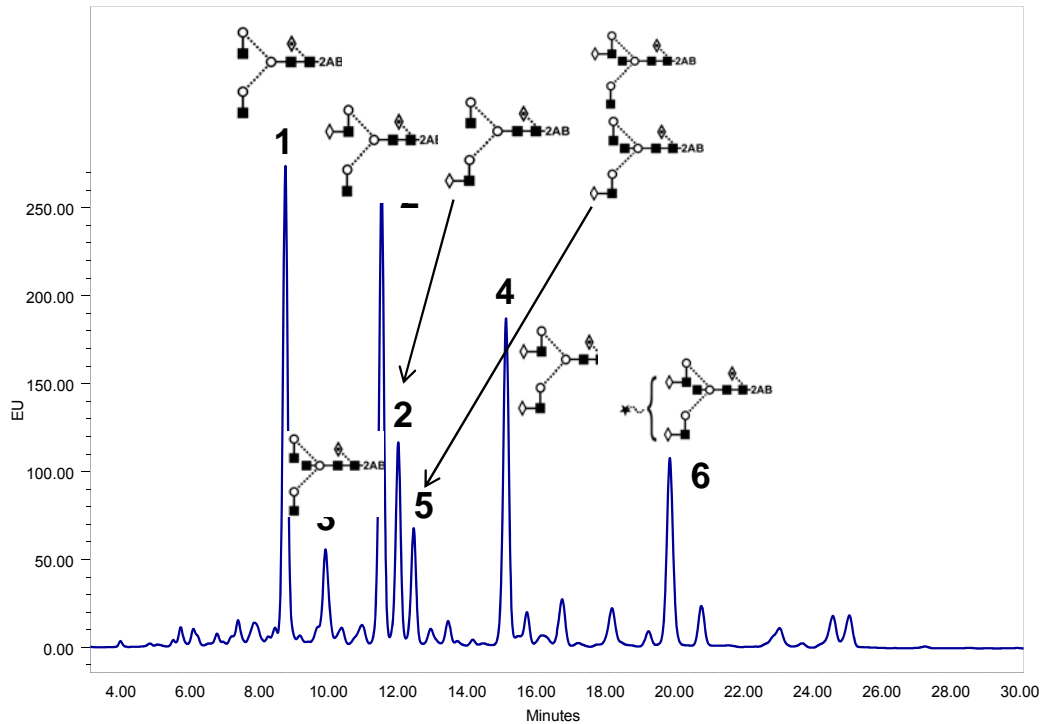


Figure 2: BEH UPLC Profile of 2AB Labelled IgG N-Glycans released from Human IgG antibody by N-mode hydrazinolysis.

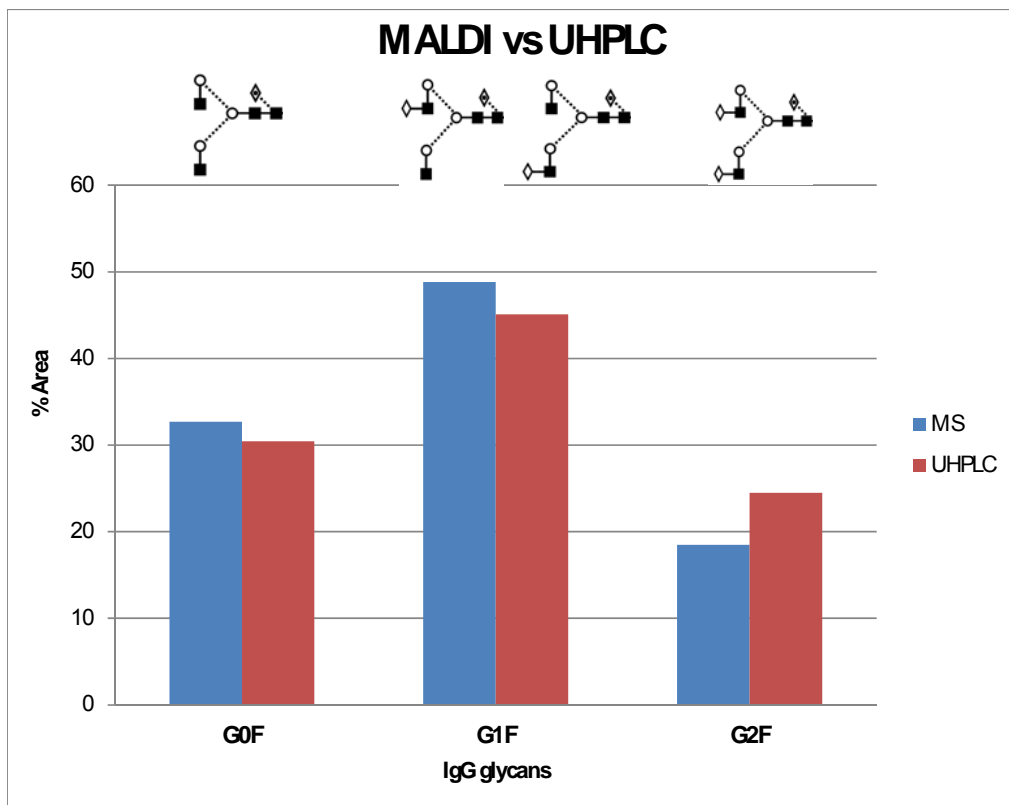
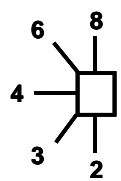


Figure 3: Comparison of MALDI MS data with Waters BEH glycan column UHPLC data for Human IgG antibody G0F, G1F and G2F structures.

Nomenclature

| <i>Symbol for sugar</i> | <i>Linkage position</i> |
|-------------------------|--|
| □ Glc |  |
| ■ GlcNAc | |
| ★ NeuNAc | |
| ◇ Gal | |
| ◆ GalNAc | |
| ◊ Fuc (deoxy galactose) | |
| ○ Man | <i>Linkage type</i> |
| | — β-linkage |
| | α-linkage |

Structure Abbreviations

All N-glycans have two core GlcNAcs; F at the start of the abbreviation indicates a core fucose, (6) after the F indicates that the fucose is α 1-6 linked to the inner GlcNAc; Mx, number (x) of mannose on core GlcNAcs; Ax, number of antenna (GlcNAc) on trimannosyl core; A2, biantennary with both GlcNAcs as α 1-2 linked; A3, triantennary with a GlcNAc linked α 1-2 to both mannose and the third GlcNAc linked α 1-4 to the α 1-3 linked mannose; A3', triantennary with a GlcNAc linked α 1-2 to both mannose and the third GlcNAc linked α 1-6 to the α 1-6 linked mannose; A4, GlcNAcs linked as A3 with additional GlcNAc α 1-6 linked to α 1-6 mannose; B, bisecting GlcNAc linked α 1-4 to α 1-3 mannose; Gx, number (x) of linked galactose on antenna, (4) or (3) after the G indicates that the Gal is α 1-4 or α 1-3 linked; [3]G1 and [6]G1 indicates that the galactose is on the antenna of the α 1-3 or α 1-6 mannose; Sx, number (x) of sialic acids linked to galactose; the numbers 3 or 6 in parentheses after S indicate whether the sialic acid is in an α 2-3 or α 2-6 linkage.