

Certificate of Stability

GPEP-IgG-01

Stability Question

Are the dried IgG glycopeptides chemically stable when subjected to mild elevated temperatures such as those that may be experienced during delivery? Glycopeptides are kept at -20°C for long term storage and delivered at ambient temperature. Users will again store the glycan at -20°C upon receipt of the product.

Stability Assay

GPEP-IgG-01 (C4BJ-02) was subjected to 37°C for a week.

Stability Outcome.

GPEP-IgG-01 (C4BJ-02) is stable after 1 week incubation at 37°C; No degradation has been observed (Figure 1-3)

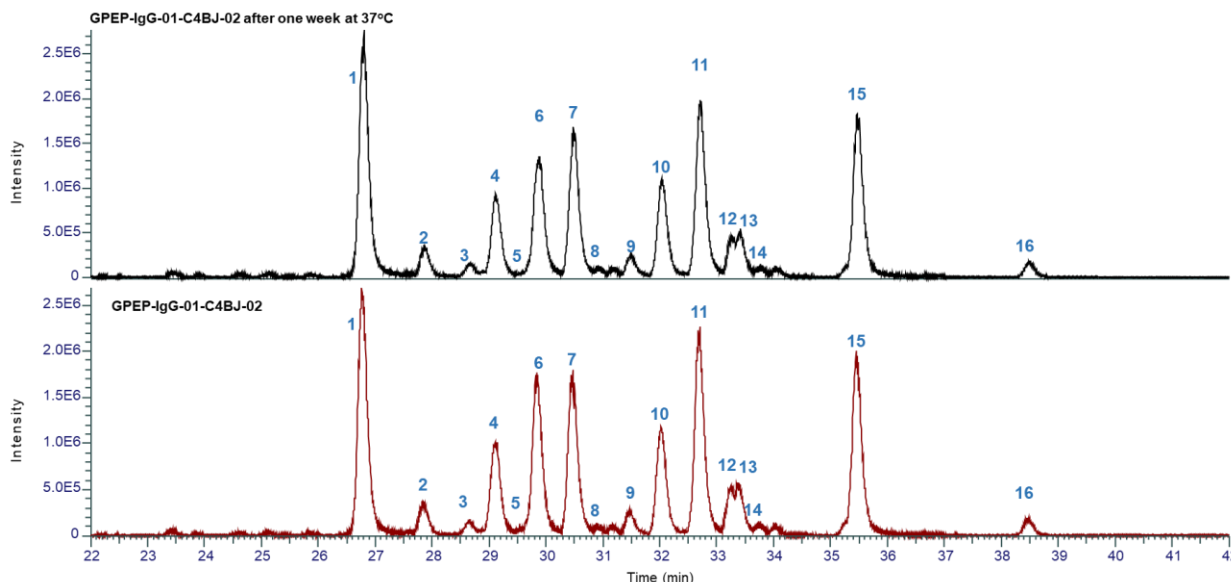


Figure 1: Base Peak chromatogram of IgG N-Glycopeptides in a HILIC-UHPLC column, digested from Human IgG antibody with Trypsin following glycopeptide enrichment. (Cat. #: CPEP-IGG-01, Batch #C4BJ-02). Top chromatogram: after being in the oven for a week at 37°C. Bottom: stored at -20°C. Table 1 shows peak assignments.

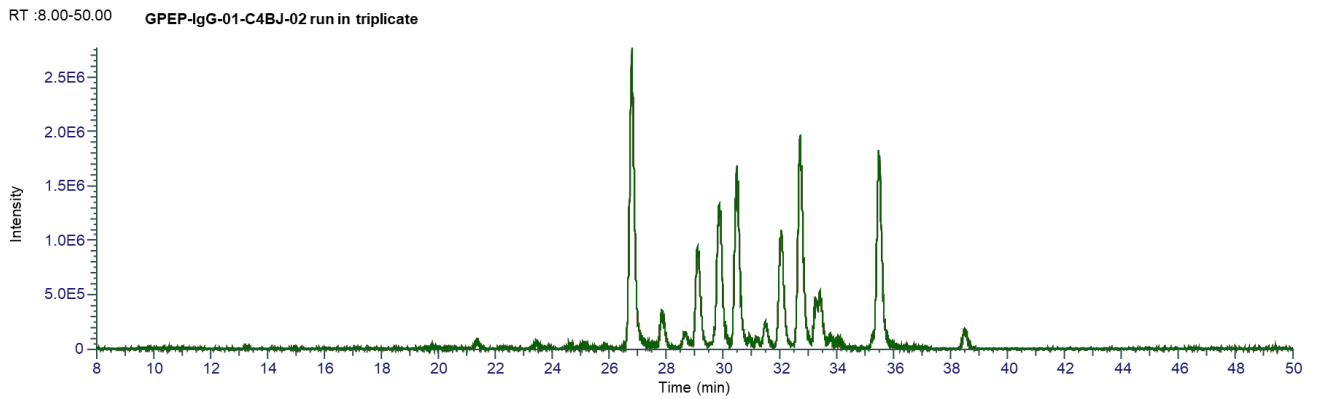


Figure 2: Base Peak chromatogram of IgG N-Glycopeptides in a HILIC-UHPLC column, digested from Human IgG antibody with Trypsin following glycopeptide enrichment. (Cat. #: CPEP-IGG-01, Batch #C4BJ-02) after being in the oven for a week at 37°C. Sample run in triplicate.

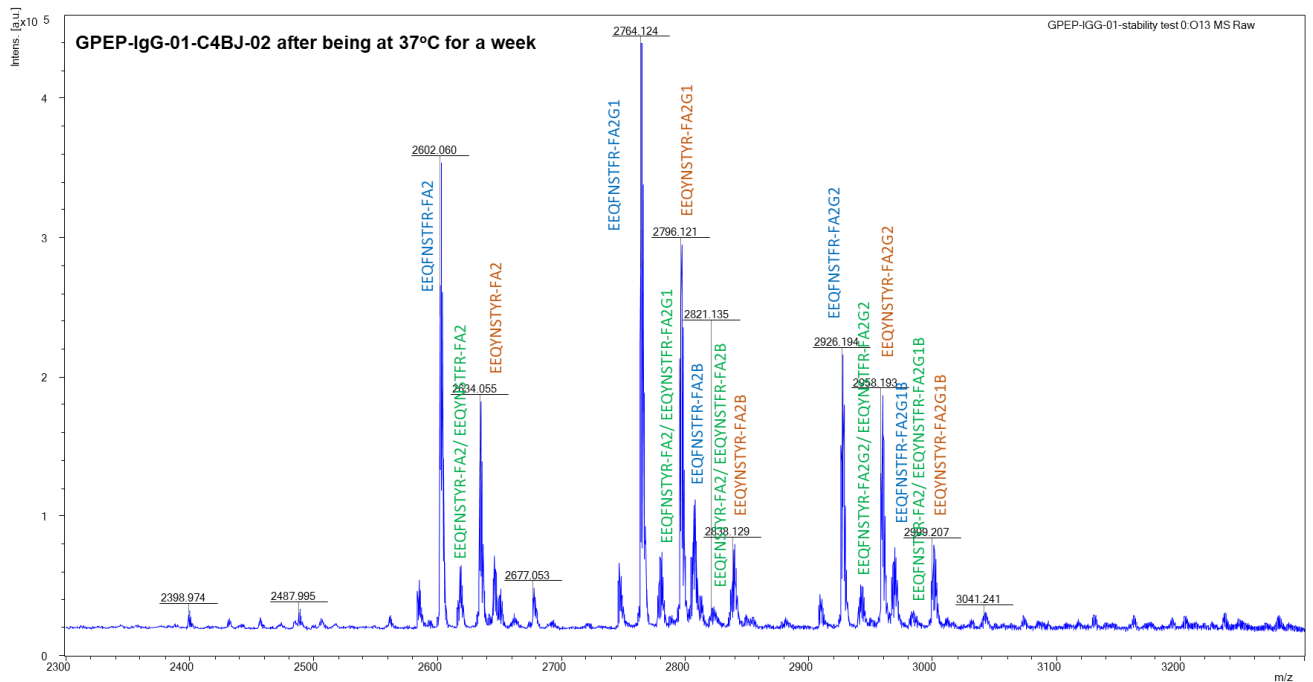













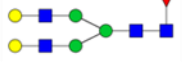
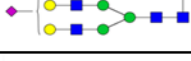



Figure 3: MALDI Mass spectrum of IgG N-Glycopeptides (Cat. #: CPEP-IGG-01, Batch #C4BJ-02) digested from Human IgG antibody with Trypsin following glycopeptide enrichment performed on Bruker Autoflex MALDI with CHCA matrix. Sample after being in the oven for a week at 37°C

Table 1: Structures and names of each peak from the BEH UPLC -ESI MS analysis

Peak ID	IgG Type	Glycopeptide name	Glycan structure
1	IgG2	EEQFNSTFR-FA2	
2	IgG2	EEQFNSTFR-FA2B	
3	IgG3/4	EEQFNSTYR-FA2/ EEQYNSTFR-FA2	
4	IgG2	EEQFNSTFR-FA2G1	
5	IgG2	EEQFNSTFR-FA2G1B	
6	IgG2	EEQFNSTFR-FA2G1	
7	IgG1	EEQYNSTYR-FA2	
8	IgG3/4	EEQFNSTYR-FA2G1/ EEQYNSTFR-FA2G1	
9	IgG1	EEQYNSTYR-FA2B	
10	IgG2	EEQFNSTFR-FA2G2	
11	IgG1	EEQYNSTYR-FA2G1	
12	IgG1	EEQYNSTYR-FA2G1B	
13	IgG1	EEQYNSTYR-FA2G1	
14	IgG3/4	EEQFNSTYR-FA2G2/ EEQYNSTFR-FA2G2	
15	IgG1	EEQYNSTYR-FA2G2	
16	IgG1	EEQYNSTYR-FA2G2S	

Structure Abbreviations

For aminoacids:

E: Glutamic acid; Q: Glutamine; F: Phenylalanine; N: Asparagine; S: Serine; T: Threonine; Y: Tyrosine and R: Arginine

For glycans:

All N-glycans have two core GlcNAcs; F at the start of the abbreviation indicates a core fucose; Ax, number of antenna (GlcNAc) on trimannosyl core: A2, biantennary with both GlcNAcs as α 1-2 linked; B, bisecting GlcNAc linked α 1-4 to α 1-3 mannose; Gx, number (x) of linked galactose on antenna; Sx, number (x) of sialic acids linked to galactose.