CERTIFICATE OF ANALYSIS

PRODUCT NAME:	AdvanceBio 2-AA Glucose Homopolymer Standard
PRODUCT CODE:	GKSA-503
LOT NUMBER:	DP14L0201
PACK SIZE:	200 pmol (qualitative standard for glycan identification)
FORM:	Dry solid
STORAGE:	Store in the dark at –20 °C before and after reconstitution
EXPIRATION:	August 2024 (extended from prior exp. date based on re-assay), may be used for 1 year after reconstitution
RE-ASSAY DATE:	August 2019
STRUCTURE:	A mixture of α (1-6)-linked glucose oligosaccharides with variable number of monomeric glucose units (1–23 or more). The reducing termini are derivatized with the fluorescent dye, 2-AA (2-aminobenzoic acid).
NOMENCLATURE:	Homopolymers of glucose derived by hydrolysis of dextran are referred to by the number of glucose monomers that each contains. Thus, glucose unit 2 is a dimer of two glucose monosaccharides and is abbreviated as GU2. This corresponds to DP2 where the “degree of polymerization” nomenclature is used as in other homopolymer series.

LABORATORY REAGENT

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Quality Control:

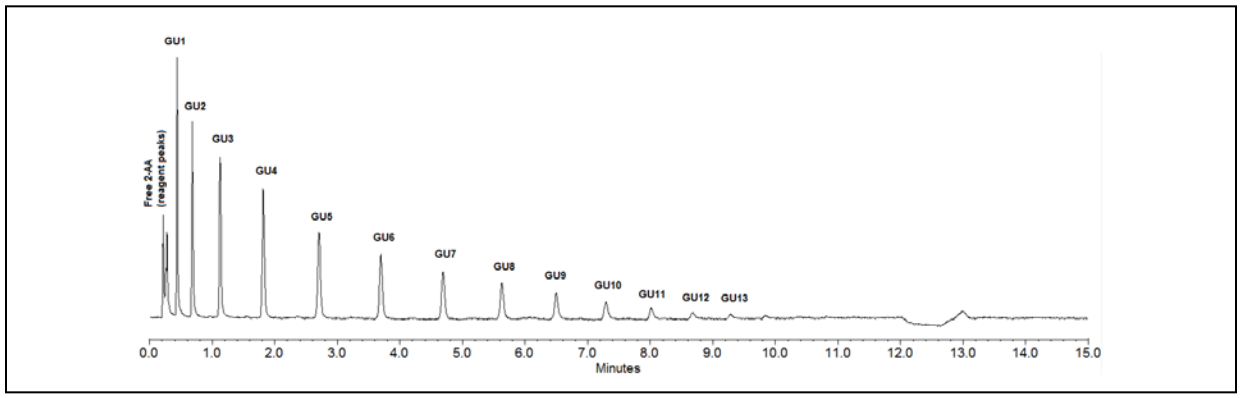


Figure 1 - **UPLC Results**: 6–9 pmol (1 µL injection) of the 2-AA-labeled[1] glucose homopolymer standard was injected on a Waters ACQUITY UPLC H Class System utilizing a 15-minute method under the conditions below:

Time (min)	Flow (mL/min)	% ACN	% Buffer
0.0	1.0	75.0	25.0
1.0	1.0	75.0	25.0
12.0	1.0	52.5	47.5
12.1	0.5	40.0	60.0
12.5	0.5	40.0	60.0
12.6	0.5	75.0	25.0
12.7	1.0	75.0	25.0
15.0	1.0	75.0	25.0

Column: Waters ACQUITY UPLC BEH Glycan Column (1.7 µm, 2.1 x 100 mm)
ACN: Acetonitrile
Buffer: 100 mM ammonium formate, pH 4.4
Flow rate: As stated in table, in mL/min
Temperature: 60 °C
Max Pressure: 15,000 psi
Fluorescence Detection: λ_{ex} = 352 nm
λ_{em} = 435 nm

Structural Analysis: UPLC Analysis of 2-AA-(Glucose Homopolymer) gives a characteristic profile with an initial non-glucose peak followed by a series of glucose polymer peaks from 1-mer to at least 23-mers. However, the number of peaks observed is dependent on the running conditions employed. The identity of the unlabeled glycan library is confirmed by MALDI-TOF mass spectrometry [2,3] or LC-MS. Agreement was found between the results from mass spectrometry and UPLC[4].

Application:

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

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

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.

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

Directions for Use: The amount of 2-AA-labeled glucose homopolymer standard injected on a UPLC column is typically 6–9 pmol of total glycan. For our Quality Control testing, one vial was dissolved in 30 µL of water and 70 µL acetonitrile with 1 µL injected on the ACQUITY column. For larger injection volumes or other LC systems we recommend further dilution as necessary for compatibility with the mobile phase.

REFERENCES

1. Bigge JC, Patel T, Bruce JA, Goulding PN, Charles SM, Parekh RB. Nonselective and efficient fluorescent labeling of glycans using 2-amino benzamide and anthranilic acid. Anal Biochem 1995 Sep 20;230(2):229-238.

2. James DC and 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.

3. Papac DI, Wong A and 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.

4. 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.

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