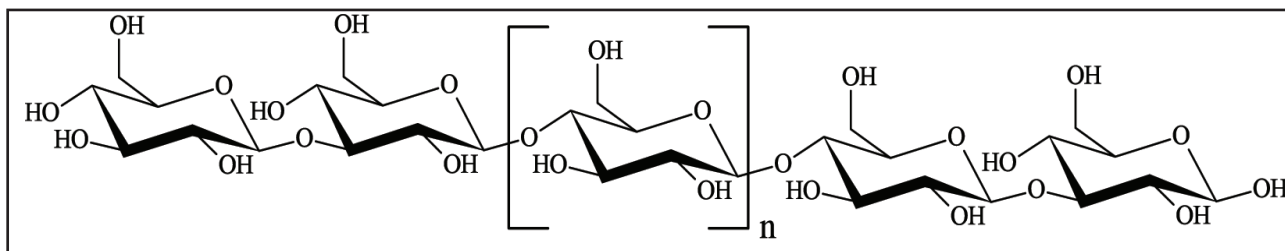# OAT BETA-GLUCAN (High Viscosity) (Lot 80608d)

P-BGOH   **04/15**
CAS NO.   9041-22-9

STRUCTURE

Schematic representation of oat β-D-Glucan subunit (n = 2 or 3; ~ 90% of glucan structure)

PROPERTIES

Purity:	> 94% (dw basis)
Viscosity:	58 cSt (1% w/v; Ostwald C-type viscometer, 30°C)
Molecular Weight:	361 Kd (MAALS)
Sugar Composition:	Glucose, 96%; (glc of alditol acetates)
Starch:	< 0.2%
Arabinoxylan:	< 0.2%
Protein:	1.4%
Moisture:	8.1%
Ash:	1.4%
Physical Description:	White, odourless powder

STORAGE CONDITIONS

Store dry at room temperature in a well sealed container. Under these conditions, the product is stable for several years.

METHOD OF DISSOLUTION (for 0.5% w/v solution)

Accurately weigh 0.5 g of β-glucan into a 120 mL dry pyrex beaker. Add 5 mL of 95% ethanol to wet the sample. Add a magnetic stirrer bar followed by 90 mL of distilled water, while stirring the slurry on a hot-plate magnetic stirrer. Adjust the heat setting to 120°C and stir vigorously. Cover the beaker loosely with aluminium foil and continue stirring vigorously. Turn the heat off when the solution begins to boil, but continue stirring the solution until the β-glucan completely dissolves (approx. 10 min). Adjust the volume of the solution to 100 mL (this solution may be very slightly turbid due to the presence of trace amounts of protein).

β-Glucan solutions can be stored at room temperature for several weeks in a well sealed storage bottle. Prevent microbial contamination by adding a few drops of toluene to the storage bottle. If the β-glucan begins to self-associate and precipitate from solution or gel, then loosen the bottle cap and heat the solution to 90-95°C in a boiling water bath for a few minutes. Tighten the cap and shake the contents vigorously. Add a few drops of toluene.

Gas liquid chromatography of the alditol acetates derived from hydrolysis and derivatisation of Oat β-Glucan (Lot 80608d)


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C-R8A CHROMATOPAC CH=1   Report No.=328    DATA=1:#CHRM1.C00   12/07/24  18:21:20
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            2.450
            3.310
            3.986
    4.0
            5.049
    6.0
            7.008
    8.0     7.842
   10.0   9.768
           10.596
   12.0
   14.0     13.588
   16.0     15.896
   18.0     17.992
   20.0                                          20.515
   22.0
   24.0
   26.0
   28.0
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GLC

A typical polysaccharide sample (~ 10 mg) was hydrolysed using 2N TFA at 120°C for 60 min. Subsequent sodium borohydride reduction was performed in 1N NH_4OH for 90 minutes at 40°C. The corresponding alditol acetates were prepared using acetic anhydride and 1-methyl imidazole, extracted into DCM and analysed by GC. Chromatography was performed on a Shimadzu GC-14B with CHROMATOPACK C-R8A using a Packed glass column (6 ft x 5 mm OD, 3 mm ID) with 3% Silar 10C on W-HP (80-100 mesh). The carrier gas was nitrogen at 130 KPa. Injector temperature; 250°C; Column temperature; 230°C. Detection by FID with 60KPa H_2 pressure and 50 KPa air pressure.

Size Exclusion Chromatography of Oat β–Glucan (Lot 80608d)
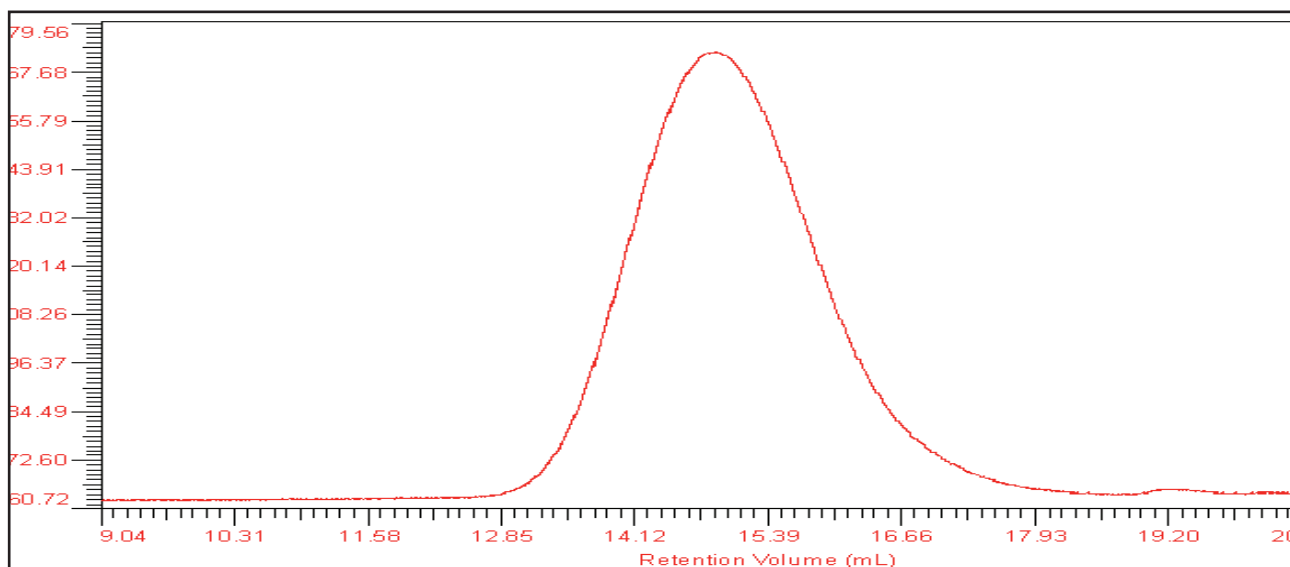
Polysaccharide & Lot Number	Mp	Mw	Mn	(η)	Rg	Pd
Oat β-Glucan Lot 80608d	305000	361000	217000	3.40	39.56	1.67
sd	10200	18200	10200	0.09	0.63	0.01

VISCOTEK ANALYSIS

Polysaccharide Solubilisation Protocol
A few mg (2-6) of the polysaccharide were weighed into a glass test tube in duplicate. Sufficient 0.1 M sodium nitrate containing 5 mM sodium azide (SEC eluent) was added to give a polysaccharide concentration of ~ 1mg/mL. The samples were stirred for 2.5 hr at 90°C. After cooling to RT, the solutions were filtered through a 0.45 μm filter into auto-sampler vials. This protocol was repeated on two separate days.

Size Exclusion Chromatography
The chromatographic system was a Shimadzu SCL-10Avp control unit (Shimadzu Scientific Instruments, Inc., Columbia, MD) using Shodex OHpak Kb-806M HQ column (Showa Denko K.K., Tokyo, Japan) followed by an Ultrahydrogel linear column (Waters, Milford, CT) maintained at 40°C and run at a flow rate of 0.6 mL/min with an eluent of 100 mM $NaNO_3$ containing 5 mM NaN_3. All measurements were made from data collected using a model 305 Triple Detector Array (TDA) from Viscotek (Viscotek, Houston, TX), which consisted of a refractive index detector, a differential pressure detector, a right angle laser light scattering detector (RALS) and a low angle laser light scattering detector (LALS). Values were calculated using OmniSEC 4.6 software (Viscotek, Houston, TX). A refractive index increment of 0.146 mL/g was used for the calculations. Pullulan standards were used to calibrate the method. For each run, two standards were used. The p100 standard was used to calibrate the method, while the p800 was treated as a sample in order to confirm the accuracy of the method. Both standards were from Fluka and were prepared in eluent.

Results
The results of the analyses are provided in the table below. The average of three determinations is reported with the standard deviation (sd).

The parameters measured are:

Mp – peak molecular weight (g/mol) – the molecular weight of the most abundant species in the sample.

Mw – weight average molecular weight – the average molecular weight of the distribution based on the weight of particles in each fraction.

Mn – number average molecular weight – the average molecular weight of the distribution based on number of particles in each fraction.

[η] – intrinsic viscosity (dL/g) – the contribution of solute molecules to solution viscosity.

Rg – radius of gyration (nm) – the root mean square distance of the monomers from the centre of the molecule.

Pd – Polydispersity Index – the ratio of Mw/Mn which is generally used as an indicator of the width of the distribution, with 1.0 representing monodisperse molecules.