Megazyme

WHEAT ARABINOXYLAN (High Viscosity) (Lot 120707a)

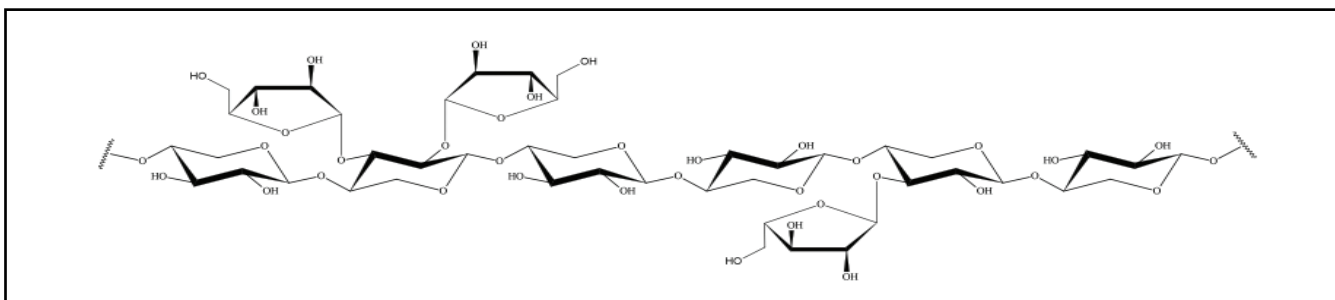**P-WAXYH**   **08/13**
CAS NO.   9040-27-1

STRUCTURE

Schematic representation of wheat arabinoxylan unit (Ara : Xyl = 38 / 62)

PROPERTIES

|	
Purity:	~ 95% (dw basis)
Sugar Ratio:	Arabinose: Xylose = 38 / 62
Viscosity:	~ 42 cSt (1% w/v; Ostwald C-type viscometer, 30°C)
Molecular Weight:	370 Kd (MAALS)
Starch Content:	0.16%
Beta-Glucan:	0.06%
Protein:	0.9%
Moisture:	3.4%
Ash:	2.8%
Physical Description:	Slightly off-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 1% w/v solution)

Accurately weigh 1 g of arabinoxylan into a 120 mL dry pyrex beaker. Wet the sample
with 8 mL of 95% ethanol. Add a magnetic stirrer bar, followed by 90 mL of distilled water.
Immediately place the beaker containing the slurry on a magnetic stirrer-hotplate and heat at a
setting of 100°C with vigorous stirring. Loosely cover the beaker with aluminium foil then stir
and boil the contents until the arabinoxylan completely dissolves (approx. 10 min). Allow the
solution to cool to room temperature with continued stirring. Adjust the volume to 100 mL.
The solution may be very slightly opalescent due to the presence of trace amounts of protein.
Arabinoxylan solutions can be stored at room temperature for several weeks in a well sealed
storage bottle. Microbial contamination is prevented by adding a few drops of toluene to the
storage bottle.

Gas liquid chromatography of the alditol acetates derived from hydrolysis and derivatisation of high viscosity wheat arabinoxylan (Lot 120707a)
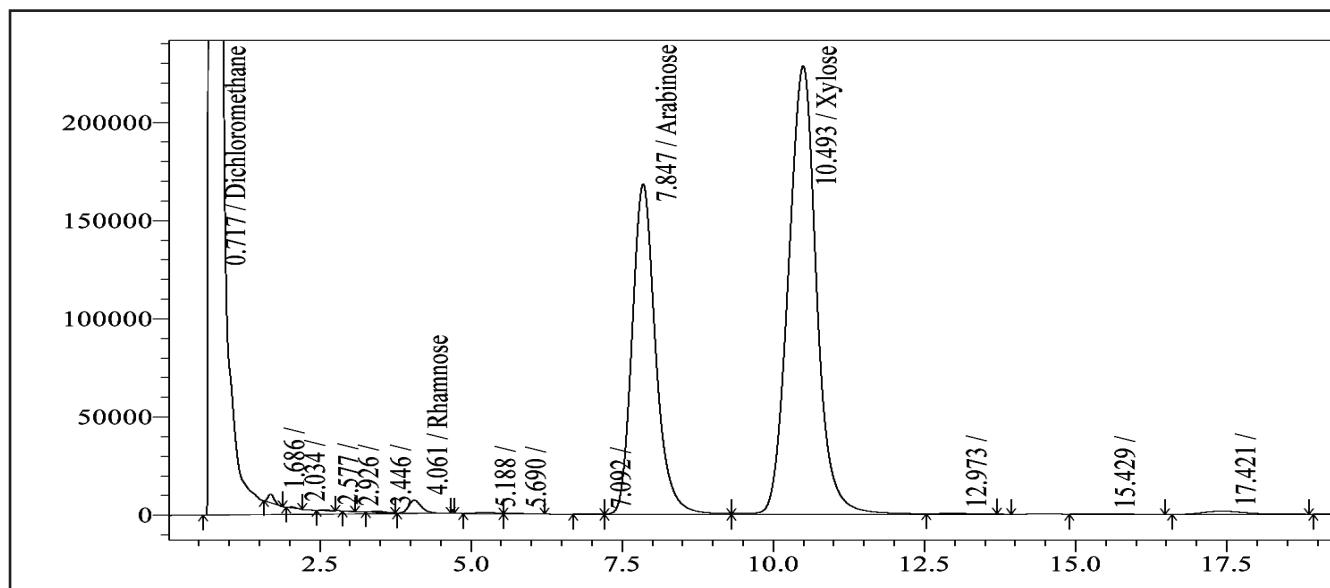
GLC

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 high viscosity wheat arabinoxylan (Lot 120707a)
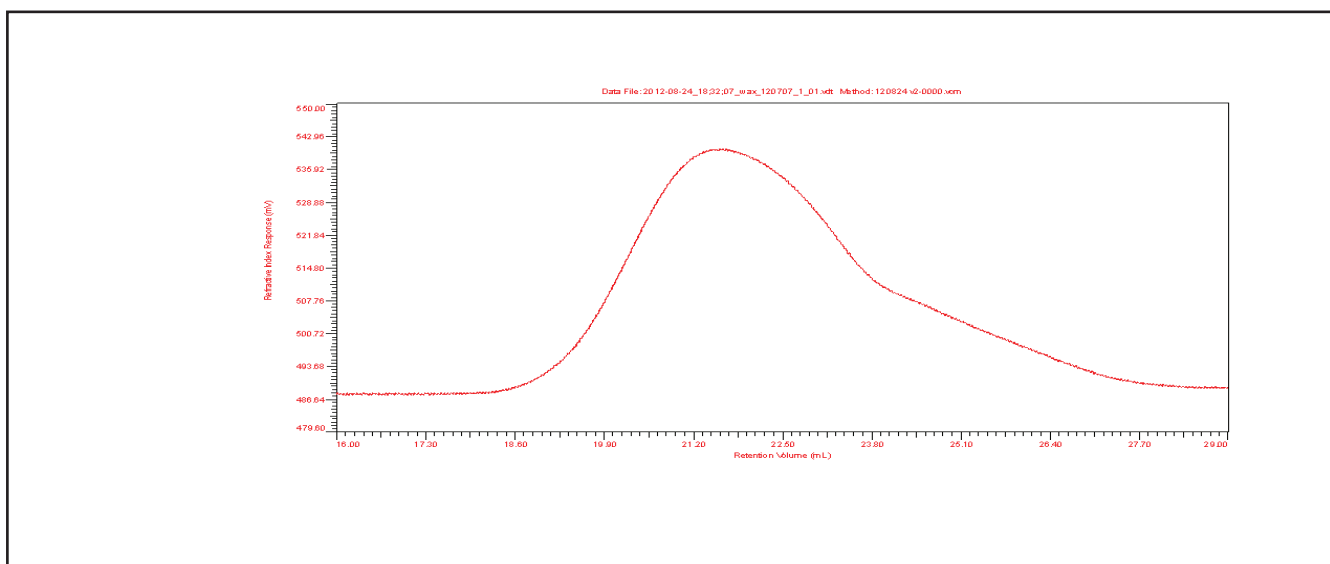
Polysaccharide & Lot Number	Mp	Mw	Mn	(η)	Rg	Pd
Wheat arabinoxylan Lot 120707	328000	370000	94000	4.30	32.4	4.11
sd	7300	2500	4900	0.05	1.3	0.44

VISCOTEK ANALYSIS

Polysaccharide Solubilisation Protocol
A few mg (2-6) of the polysaccharide were weighed into a glass test tube in triplicate. 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 two PAA-M mixed bed columns and 1 PAA-203 column, plus a guard (PolyAnalytik, London, ON, Cd.) 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 from Viscotek (Viscotek, Houston, TX), which consisted of a refractive index detector (RI), a differential pressure detector (DP), 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.