Megazyme Building 1,
Bray Business Park,
Bray, Co. Wicklow,
A98 YV29,
Ireland.
Tel: + 353 1 2861220 Fax: + 353 1 2861264

Certificate of Analysis

Product: Arabinoxylan (Rye Flour)
Cat. No: P-RAXY
Lot. No: 151208
Expiry Date: N/A

Parameter	Specification	Results
Purity	> 90% (dw basis)	90%
Viscosity	70 – 100 cSt	95 cSt*
Starch	< 0.1%	0.04%
Beta Glucan	< 0.1%	0.06%
Protein	< 5%	3.3%
Moisture	< 2%	1.9%
Ash	< 10%	5.3%

*1% w/v solution, 30°C, Ostwald C-type viscometer.

Sugar Composition:

Arabinose	40%
Xylose	60%

This lot meets our specification.

Signature: Ciara McLoughlin **Date:** 02.Feb.2022

Ciara McLoughlin
QC Specialist

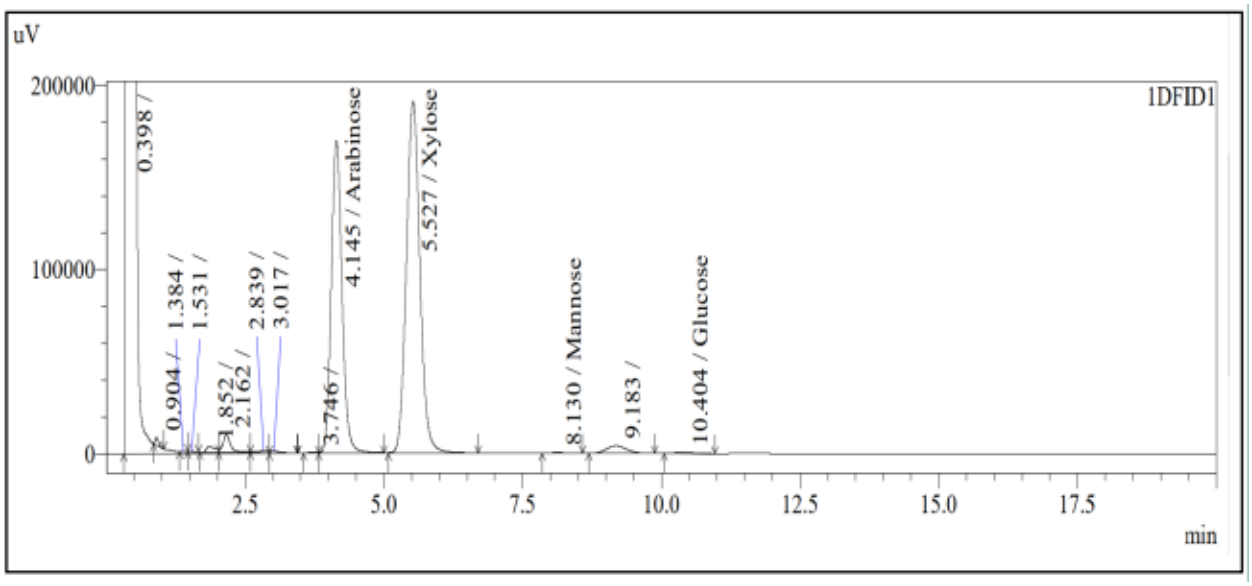
(*as this certificate is a computer printout, it has not been signed or dated by hand*)

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GC:

Gas liquid chromatography of the alditol acetates derived from hydrolysis and derivatization of High viscosity Rya Arabinoxylan (Lot 151208)



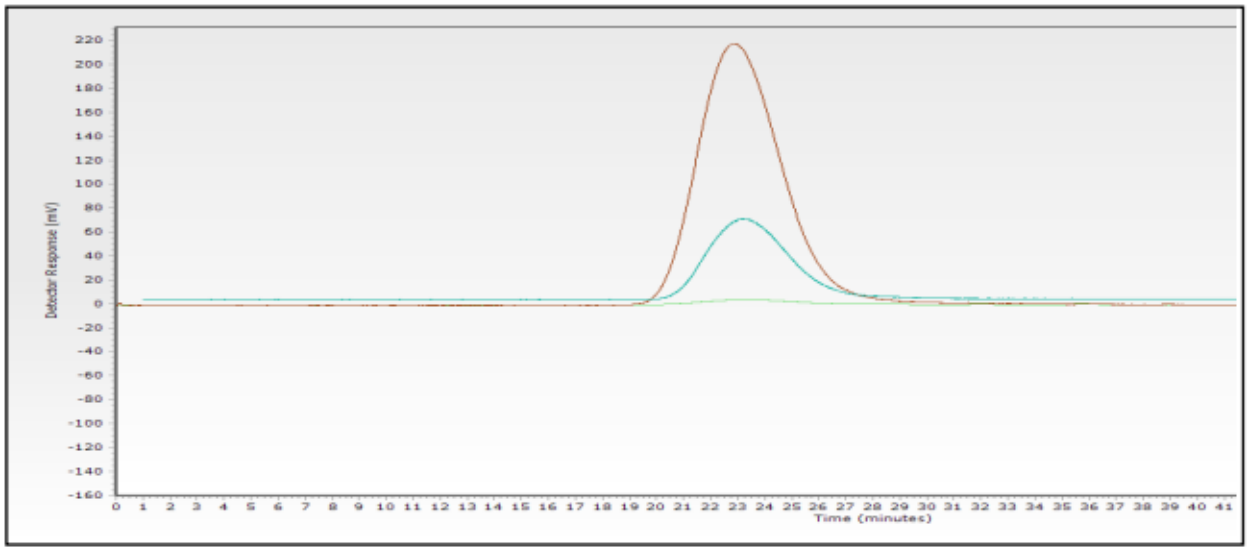
GLC:

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 NH4OH 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 (6ft x 5 mm OD, 3mm 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 H2 pressure and 50 KPa air pressure.

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Size Exclusion Chromatography of high viscosity wheat arabinoxylan (Lot 151208)



Polysaccharide & Lot Number	Mp	Mw	Mn	Pd
Rye Arabinoxylan Lot 151208	350897	385986	197912	1.83

Viscotek Analysis

Polysaccharide Solubilisation Protocol

10 mg of the polysaccharide were weighed into a glass test tube. 10mL 0.1 M $NaNO_3$ containing 5 mM NaN_3 (GPC-SEC eluent) was added to give a polysaccharide concentration of ~ 1 mg/mL. The samples were sealed and stirred for 2.5 h at 90°C. After cooling to RT, the solutions were filtered through a 0.2 µm filter and analysed. This protocol was repeated on two separate samples.

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Size Exclusion Chromatography

GPC/SEC chromatography was performed on an Agilent 1260 Infinity using a Shodex OHpak SB-806M HQ column (8 x 300 mm) followed by an Ultrahydrogel linear column (7.8 x 300 mm) maintained at 40°C using an eluent of 0.1 M $NaNO_3$ containing 5 mM NaN_3 and flow rate of 0.6 mL/min. An Infinity 1260 Triple Detector Suite from Agilent Technologies was used, consisting of a refractive index (RI) detector, a viscometer (VS) detector and a dual angle (90° and 15°) laser light scattering (LS) detector. Triple detection analysis was done using the Agilent GPC/SEC software (Version A:02:01). A refractive index increment (dn/dc) of 0.146 mL/g was used for the calculations.

Results

The results of the analyses are provided in the table below.

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.
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.

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