ELSEVIER

Carbohydrate Research 281 (1996) 161–172

CARBOHYDRATE RESEARCH

Structural features of an arabinan fragment isolated from the water-soluble fraction of dehulled rapeseed

Ingrid Eriksson [a,*], Roger Andersson [a], Eric Westerlund [a], Rolf Andersson [b], Per Åman [a]

[a] *Department of Food Science, Swedish University of Agricultural Sciences, P.O. Box 7051, S-750 07 Uppsala, Sweden*

[b] *Department of Chemistry, Swedish University of Agricultural Sciences, P.O. Box 7051, S-750 07 Uppsala, Sweden*

Received 27 April 1995; accepted 15 September 1995

Abstract

A water-soluble polysaccharide fraction was prepared from dehulled rapeseed meal (winter rapeseed variety Casino). Further purification yielded two major fractions having a high content of arabinose and galactose residues, with Ara/Gal ratios of 5.4 (G1) and 1.8 (G2). The Ara/Gal ratio of the high molecular weight fraction G1 was stable over the whole gel filtration peak, indicating that the arabinose and galactose residues are part of the same polysaccharide. The high molecular weight fraction G1 was studied further by methylation analysis and several NMR techniques. Structural studies showed G1 to consist mainly of arabinan fragments, which have terminal α-L-arabinofuranosyl groups with anomeric carbons bound (1 → 5) (A) or (1 → 2) (B), and 2,5-substituted arabinosyl residues with anomeric carbons bound (1 → 5) (D) or (1 → 2) (C) to adjacent arabinosyl residues. The A:B:C:D ratios were 2:1:1:1 according to results from NMR and methylation analysis.

Keywords: Rapeseed; Arabinan; NMR

1. Introduction

Arabinans can be found in various plant materials, notably in seeds, fruits, and roots. Isolation and investigation of arabinan structures from cotyledons of red gram [1], roots of marsh mallow [2], roots of horsebean [3], cabbage [4], grape juice [5], apple juice [6],

* Corresponding author.

SSDI 0008-6215(95)00337-1

and inner bark of *Rosa glauca* stems [7] have been described, to name a few. These arabinans, whether occurring as sidechains to pectins or as polymers associated with pectin, mostly consist of (1 → 5)-linked α-L-arabinofuranosyl residues. The structural investigation of these isolates has mainly involved methylation analysis and ^{13}C NMR spectroscopy. Arabinogalactans and arabinogalactan-proteins are also widely distributed in higher plants [8,9]. In the 1970s, two groups studied rapeseed polysaccharides, including arabinan and arabinogalactan structures, mainly using methylation analysis as a tool for structural analysis [10–15]. Since then, rapeseed polysaccharide work has focused on the content and composition of the dietary fibre fraction, rather than on structural analysis of isolated polysaccharides. The present paper will, however, describe structural features of a water-soluble rapeseed polysaccharide, containing arabinan fragments, on the basis of methylation analysis and several NMR techniques. This knowledge might be useful to the further understanding of the nutritional and technological properties of rapeseed.

2. Experimental

General analyses.—The analyses were made at least in duplicate and the results are reported on a dry-matter basis. Defatting of samples prior to fibre analyses was done according to Eriksson et al. [16]. Analyses of dietary fibre components in whole seed and fractions of rapeseed were made according to the Uppsala method of dietary fibre determination [17], modified as described below. The starch degradation step in the method was excluded, because of the insignificant amount of starch in the samples. Also, Klason lignin was determined without ashing. Uronic acid values were corrected with a factor of 0.88 instead of 0.81. Incubation with concentrated sulfuric acid in the hydrolysis step was excluded when analysing the water-soluble fraction.

The neutral polysaccharide content in gel filtration fractions was determined by sugar analysis including hydrolysis in 1 M trifluoroacetic acid (90 min, 125 °C), reduction, and acetylation using 1-methylimidazole as a catalyst. The resulting alditol acetates were quantified by GLC as in the Uppsala method.

Preparation of meal with low hull content.—Samples of mature seeds (3 × 250 g), from the winter rapeseed variety Casino, were ground in a coffee-type mill (Janke and Kunkel, IKAWERK, Germany) to a particle size of less than 0.5 mm. After extraction, twice with hexane (500 mL) in an ultrasonic bath (10 min) at room temperature, and centrifugation (1000 *g*, 10 min), the pellets were refluxed (30 min) in aq 80% EtOH (500 mL) in a boiling-water bath. The samples were then centrifuged (1000 *g*, 10 min), and the pellets extracted by ultrasonication as above and dried at 40 °C under reduced pressure. The resulting meal samples were ground in a mortar and suspended in hexane at a sample–hexane ratio of 1:5. This was done in order to separate the hulls from the rest of the meal, utilising the different density of the two components. This method of dehulling rapeseed has been reported on an industrial scale using a liquid cyclone [18]. Consequently, centrifugation (1000 *g*, 10 min) yielded a lower layer of hull-enriched meal and an upper layer of meal with low hull content. Separation of the layers was done manually, after decantation of hexane, and the resulting fractions were dried at

room temperature overnight. The replicates obtained were analysed for content of Klason lignin and polysaccharide residues; on the basis of these results, the fractions of low hull content (denoted as dehulled meal) were pooled.

Isolation of water-soluble polysaccharides.—Water-soluble polysaccharides were prepared from duplicate samples of dehulled meal (2 × 50 g), essentially as described for isolation of water-soluble mixed-linked β-glucans in oat milling fractions [19]. Each sample was divided into six portions of 8–9 g, which were extracted (by vigorous shaking) four times with 2:3 2-propanol–hexane (100 mL) and three times with aq 80% EtOH (100 mL). Each extraction was followed by centrifugation (1500 *g*, 15 min) and decantation of supernatant solutions. The final pellets were then pooled into a 1-L Duran bottle, and preheated water (750 mL, 96 °C) containing 375 μL of Termamyl 300 L (Novo Nordisk A/S, Denmark) was added. The mixture was incubated (2 h, 96 °C) in a water bath with shaker and then centrifuged (1500 *g*, 30 min), the supernatant decanted, and the pellet washed by centrifugation with water (2 × 25 mL). The supernatant solutions were then pooled into a 1-L Duran bottle and incubated (3 h, 40 °C) with NaN_3 (160 mg) and pancreatine (30 mg; Sigma P-7545, St Louis, USA) in a water bath with shaker. Enzymes were then inactivated by boiling for 20 min and the ca. 800 mL was concentrated to ca. 200 mL in a rotary evaporator at 40 °C before dialysis (Spectra/por; molecular weight cut-off, 12–14,000) against deionised water for at least 72 h. The sample was then centrifuged (1500 *g*, 30 min) and floating particles in the supernatant solution were removed by filtration through a glass filter funnel (porosity grade 3) before freeze-drying. The water-soluble and insoluble fractions were analysed for content of polysaccharide residues and Klason lignin.

Gel filtration.—Part of the water-soluble fraction (5 mg) was suspended in 0.5 M aq NaCl and centrifuged (1500 *g*, 20 min), and the supernatant solution was fractionated in a small-scale test-run on a Fractogel TSK HW-75(S) column (30 × 1.6 cm) in a Pharmacia FPLC system. The column was calibrated with D-glucose and dextran T 2000. Elution with 0.5 M NaCl (0.3 mL/min) was followed by refractive index (RI) and UV (254 nm). Fractions of 1 mL were collected and subjected to sugar analysis.

For preparative purposes, part of the water-soluble fraction (2 × 50 mg) was dissolved in 0.5 M NaCl and applied on a Sephadex G 150 column (90 × 2.6 cm). Elution with 0.5 M NaCl (0.5 mL/min) was monitored by RI and UV (254 nm), and fractions of 8 mL were collected. A high molecular weight polysaccharide fraction was obtained by pooling subfractions according to their RI and content of neutral polysaccharide residues, determined by sugar analysis. The pooled material was dialysed as above and freeze-dried before further purification by repeated use (10 runs) of the Fractogel TSK HW-75(S) column. Two fractions, G1 (elution volume, 29–39 mL) and G2 (40–49 mL), were collected, dialysed, and freeze-dried before further analysis.

Methylation analysis and GLC-MS.—Samples of fraction G1 (0.5 mg) were methylated essentially according to Harris et al. [20], in several steps due to difficulties of dissolving in Me_2SO. Methylated samples were hydrolysed in 1 M trifluoroacetic acid (90 min, 125 °C), reduced, and acetylated using 1-methylimidazole as a catalyst. GLC-MS was performed with an HP 5890 gas chromatograph, equipped with a DB-1 capillary column and connected to a Trio-1 (Fisons Instruments) mass spectrometer. The oven temperature was programmed from 150 °C (held for 2 min) to 225 °C at 2 or 3

°C/min. The injector temperature was 240 °C and the inlet helium pressure was 75 kPa. EI-spectra were recorded with the ion source energy set at 70 eV. Identifications were made by comparison with standards, and published mass spectral data [21] and retention times [22]. Quantification was made by GLC, with a flame-ionisation detector, under the same conditions as used above. Response factors, according to the effective carbon response theory [23], were applied to integrated areas.

NMR spectroscopy.—Fraction G1 was characterised by several different NMR methods, using standard pulse sequences. Spectra were recorded either on a Varian VXR 400 or a Varian Unity 600 instrument, at 30 °C using D_2O as solvent. 1D ^1H NMR spectra were also recorded at 85 °C. Chemical shifts were measured in relation to internal dioxane (67.4 ppm, ^{13}C NMR) or internal sodium 4,4-dimethyl-4-sila(2,2,3,3-$^2H_4$)pentanoate (0 ppm, ^1H NMR). The phase-sensitive ^1H-detected $^1H,^{13}C$ HMQC (heteronuclear multiple quantum coherence) spectrum (600 MHz) was recorded with 32 transients over 512 increments (zero-filled to 1K) and 1K data points with spectral widths of 3380 Hz in F_2 and 10,000 in F_1. The repetition time was 2.3 s. The delays were adjusted according to a coupling constant ^1J(CH) of 145 Hz. Gaussian apodisation was applied in both dimensions. The phase-sensitive 600-MHz NOESY (nuclear Overhauser effect spectroscopy) experiment was recorded with 180 increments (zero-filled to 1K) and 1K data points. The spectral width was 3380 Hz in both dimensions; repetition time, 2.3 s; and mixing time, 0.15 ms. Gaussian apodisation was applied in both dimensions. The 600-MHz $^1H,^1H$ TOCSY (total correlation spectroscopy) experiments were recorded under similar conditions as the NOESY experiment with mixing times of 10, 30, 50, and 70 ms. The broad-band decoupled ^{13}C spectrum (101 MHz) was recorded with 16,000 data points; spectral width, 20,000 Hz; repetition time, 0.9 s; and a pulse angle of 45°. Line broadening of 3 Hz was applied.

Integration of anomeric arabinose peaks in the 600-MHz ^1H NMR spectrum was performed with the aid of symmetric Lorentzian curve-fitting in the NMR1™ NMR processing software version V1R4V1 (New Methods Research Inc., East Syracuse, NY, USA).

3. Results and discussion

It has been established earlier that the dietary fibre compositions of rapeseed hull and dehulled seed are quite different [16]. The content of Klason lignin and uronic acid was found to be relatively higher in the hull, whereas that of arabinose residues was higher in the dehulled seed. To obtain more knowledge of the composition and structure of the cotyledon polysaccharides, it was deemed neccessary to obtain a meal as hull-free as possible. Three fractions of meal with low hull content were prepared from the winter rapeseed variety Casino, extensively grown in Sweden. The hull content of these fractions was estimated to be low by comparing their Klason lignin values to those of manually dehulled seed. This was possible because Klason lignin values for rapeseed are highly correlated to the hull content [16]. From these pooled, virtually hull-free fractions (yield: 32% defatted, dehulled meal from defatted whole seed), a water-soluble fraction was obtained, constituting 2.9% of the defatted dehulled meal. Similar yields of

Table 1
Content of polysaccharide residues and Klason lignin (Kl) in different fractions prepared from the winter rapeseed variety Casino, expressed as percentage of fraction, dry weight

Fraction	Rha	Fuc	Ara	Xyl	Man	Gal	Glc	Uronic acids	Kl	Sum
Whole seed	0.23	0.13	2.86	1.05	0.31	1.29	4.18	3.90	4.2	18.18
Hull [a,b]	0.81	0.28	7.77	1.76	1.00	3.46	15.15	18.84	28.46	77.48
Dehulled seed [a,b]	0.50	0.30	5.13	2.46	0.56	2.50	7.30	3.95	1.28	23.97
Dehulled meal [b]	0.50	0.31	6.36	2.88	0.59	2.86	8.25	4.89	1.28	27.91
Water-soluble fraction	0.54	0.43	26.72	2.97	1.89	11.4	5.12	5.57	–	54.64
Gelfiltration fractions										
G1	–	–	73.2	3.1	1.4	13.6	3.1	2.4	–	96.8
G2	0.7	0.5	44.7	5.8	0.9	24.3	9.5	10.5	–	96.8

[a] Manually dehulled.
[b] Percentage of fraction when defatted and extracted with hexane, 80% ethanol, and acetone according to Eriksson et al. [16].

water-soluble polysaccharides from rapeseed have been obtained earlier [24,25]. Gel filtration of the water-soluble fraction, described further below, yielded two main fractions: G1 (9% of the water-soluble fraction) and G2 (10%). The polysaccharide compositions of these fractions, whole seed, manually dehulled seed, and hull are listed in Table 1. Values for dehulled meal, manually dehulled seed, and hull are expressed as the percentage of sample defatted according to Eriksson et al. [16]. It was notable that arabinose and galactose residues were enriched in the water-soluble fraction compared to the dehulled meal, while especially the content of glucose residues was lower. This was even more apparent in the fraction G1, where the content of arabinose residues was especially high.

Gel filtration.—Gel filtration of the water-soluble fraction was first done in a test-run on a Fractogel TSK HW-75(S) column and it was shown by sugar analysis (Fig. 1) as well as RI detection that the material was mainly eluted in two peaks. The corresponding UV-curve (254 nm) indicated that the second peak also contained large amounts of protein or other substances containing aromatic groups. It can be seen that the polysaccharide composition of the first peak was fairly constant over the whole peak, illustrated by the Ara/Gal ratio curve shown in Fig. 1. This suggests that both the arabinose and galactose residues are part of the same polysaccharide. Small amounts of glucose and xylose present could be due to xyloglucans contaminating the fraction. The second peak, on the other hand, contained high amounts of arabinose and galactose, but other sugars were also notably present. Also, the optima of the individual sugar residue peaks did not coincide completely (in the case of xylose and mannose, for example). These observations indicate that the second peak is a mixture of at least 2 polysaccharides, probably including an arabinogalactan and a xyloglucan. For preparative purposes, 100 mg of the water-soluble fraction was first applied on a Sephadex G 150 column and ~ 34 mg from a peak of higher molecular weight was recovered, whereas material of lower molecular weight, containing the UV-absorbing compounds, was discarded. The recovered high molecular weight material was fractionated on the Fractogel TSK HW-75(S) column, as

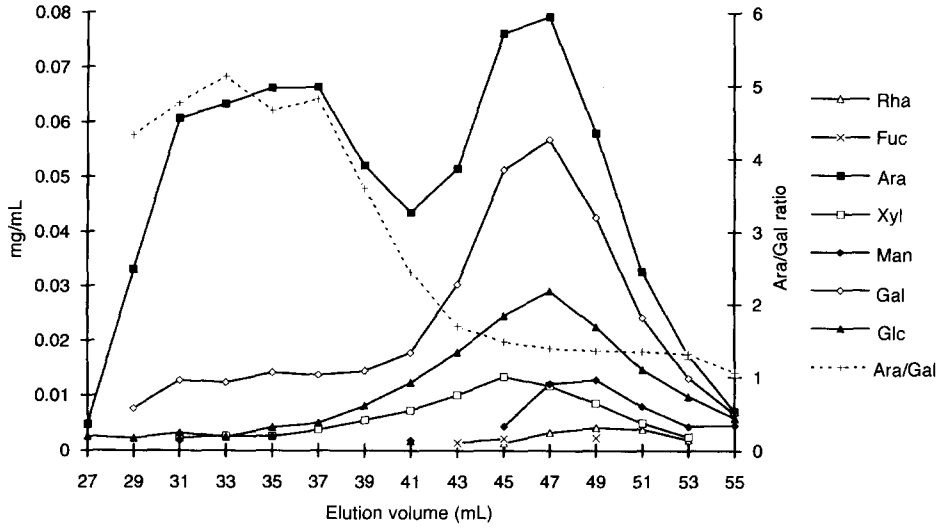

Fig. 1. Gel filtration of water-soluble polysaccharides from rapeseed on Fractogel TSK HW-75(S) eluted by 0.5 M aq NaCl. Elution was followed by sugar analysis of 1-mL fractions and the contents of individual sugar residues are expressed as mg/1-mL fraction. Elution volume of the void was ~ 25 mL and that of glucose (included volume) ~ 56 mL.

described above, and fractions G1 (elution volume, 29–39 mL) and G2 (40–49 mL) were collected on the basis of RI-detection and sugar analyses from the test run. The molecular weights of the fractions were not determined specifically, but the molecular weight range for the gel medium employed was high (10^5–10^7 according to dextran standards). Therefore, the molecular weight of G1 in particular can be assumed to be high, since this fraction was eluted fairly early after the column void volume of 25 mL.

Structural investigation.—The proportions of the different methylated derivatives in fraction G1 are given in Table 2. In agreement with sugar analysis, arabinose was the predominant sugar, mainly present as terminal (45.7%) and 2,5-substituted arabinosyl residues (34.6%). Unlike most arabinans, there was a low proportion of 5-substituted arabinose (4.0%). Galactose was mainly present as 3,6-substituted (6.1%) and, to a lesser extent, 3-substituted galactose residues (1.4%). These data indicate that the polysaccharide is highly branched and that the arabinose residues, terminal or otherwise, are mainly (1 → 2)- and (1 → 5)-linked. The result obtained from the methylation analysis was similar to that found in a previous study [10], where a tentative structure of a highly branched arabinogalactan was presented. To obtain more information on how the individual residues were linked in the fraction G1, several NMR methods were employed, as described in the Experimental section. It was possible to assign most of the signals of the ^{13}C NMR spectrum (Fig. 2) by comparison with previously published ^{13}C-spectral data on arabinans [1–3,7], and the ^{13}C chemical shifts of methyl *O*-methyl-α-D-arabinofuranosides [26]. The signals at 108.3, 77.4, and 62.0 ppm thus correspond to C-1, free C-3, and free C-5, respectively, of terminal arabinosyl groups. The complex set of signals centred at 67.1 ppm most probably refers to C-5 of 2,5-substituted

Table 2
Methylation analysis data on the water-soluble polysaccharide fraction G1 from the winter rapeseed variety Casino, obtained by gel filtration

Sugar derivative [a]	Mole (%)
2,3,5-Me_3-Ara [b]	45.7
2,3,4-Me_3-Pentitol	2.2
2,3-Me_2-Ara	4.0
2-Me-Ara	0.5
3-Me-Ara	34.6
Ara	3.9
2,4,6-Me_3-Gal	1.4
2,4-Me_2-Gal	6.1
2-Me-Gal	1.1
Gal	0.5

[a] Individual sugar derivatives are not included when constituting less than 0.5% of the total.
[b] 2,3,5-Me_3-Ara = 1,4-di-*O*-acetyl-2,3,5-tri-*O*-methylarabinitol, etc.

arabinosyl residues, in various chemical environments. This would agree with the corresponding C-3 signal at 76.1, C-2 at 88.5, and C-1 at 107.2 ppm, and is also supported by the large proportion of 2,5-substituted arabinosyl residues found by methylation analysis. The remaining C-4 and C-2 signals are more difficult to assign since they partly overlap. It is most probable, however, that the signals in the range 81.6–83.6 ppm can be attributed to C-2 of terminal arabinosyl groups, overlapping with C-4 of 2,5-substituted arabinose, and that the signals in the range 84.4–85.2 ppm correspond to C-4 of terminal arabinosyl groups. Furthermore, the chemical shifts in the ^{13}C spectrum agree with previous data for arabinans, to indicate that the arabinosyl

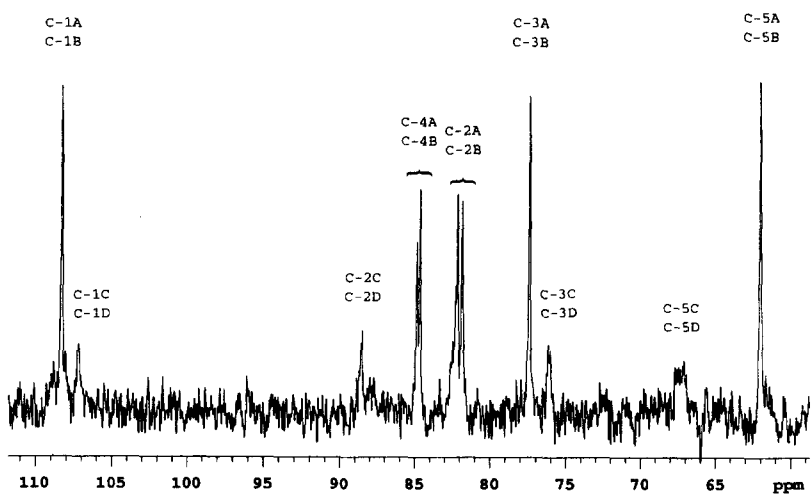

Fig. 2. 101-MHz ^{13}C NMR spectrum of fraction G1 recorded at 30 °C with D_2O as solvent. The letters A, B, C, and D in the spectrum refer to corresponding residues in the structure (see Fig. 6).

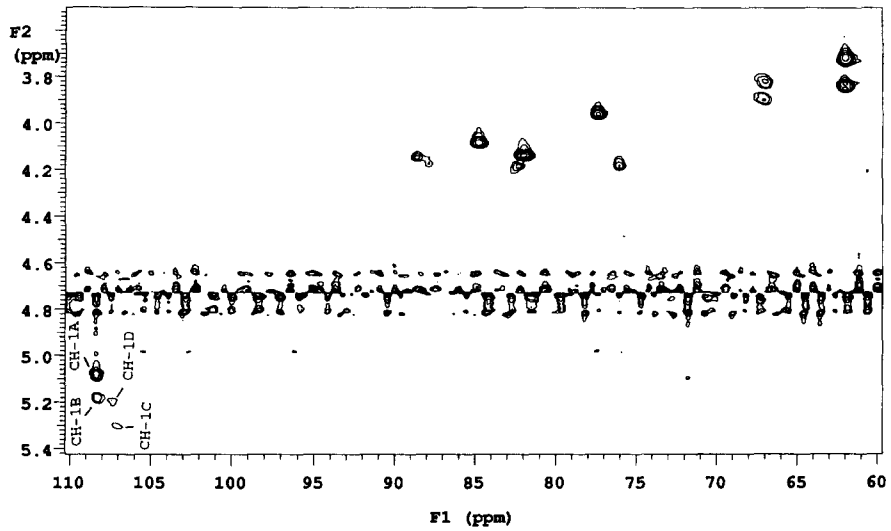

Fig. 3. 600-MHz $^{1}H,^{13}C$ HMQC spectrum of fraction G1 recorded at 30 °C with D_2O as solvent. The letters A, B, C, and D in the spectrum refer to corresponding residues in the structure (see Fig. 6).

residues are α-furanoid [1–3,7]. In addition, arabinosyl residues in water-soluble arabinan and arabinogalactan isolated from rapeseed have been reported to be α-L-furanoid [10,11].

Cross-peaks between ^{13}C and ^{1}H obtained in the HMQC experiment were used to identify signals in the ^{1}H spectrum. Unfortunately, several of the peaks overlap, especially those for H-2, H-3, and H-4. The assignments that could be made are shown in Fig. 3. This overlapping of peaks together with the small coupling constant between H-1 and H-2 in arabinose also made it difficult to interpret the $^{1}H,^{1}H$ TOCSY (total correlation spectroscopy) experiments run in the course of this investigation. However, with combined information from HMQC and NOESY spectra, the arabinose anomeric signals in the ^{1}H spectrum (Fig. 4) could be identified. The large C-1 signal at a chemical shift of 108.3 ppm from terminal arabinosyl groups thus refers to two anomeric proton peaks in the ^{1}H spectrum (at 5.08 and 5.18 ppm) as follows from the corresponding cross-peaks in the HMQC spectrum (Fig. 3). Likewise, the ^{13}C signal at 107.2 gave two cross-peaks, at 5.20 and 5.30 ppm respectively. This indicates that two main types of arabinosyl residue are present in the polysaccharide, each bound via the anomeric carbon in two different ways (at least), either (1 → 5), (1 → 2), or (1 → 3). However, the (1 → 3) alternative could be ruled out, since no signal corresponding to glycosidically bound C-3 was detected in the ^{13}C spectrum and results from methylation analysis indicated very few arabinose residues (4.4%) bound through O-3. In the NOESY spectrum (Fig. 5), cross-peaks from the anomeric protons to other parts of the spectrum indicate the presence of inter-residue bonds between arabinose residues. Consequently, the NOESY experiment showed that the large, sharp H-1 signal at 5.08 ppm corresponds to a terminal arabinosyl group (residue A, Fig. 6), glycosidically bound (1 → 5) to an adjacent arabinosyl residue, because of the large cross-peaks to the H-5 region of the

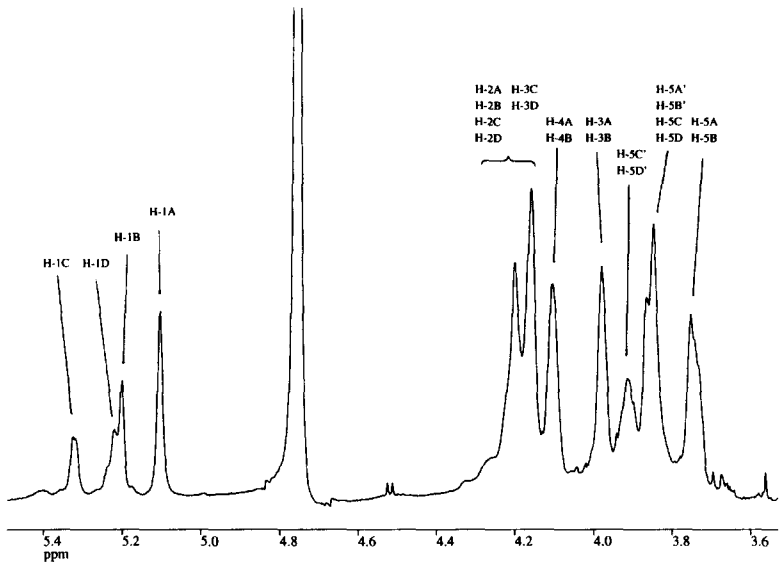

Fig. 4. 600-MHz ^{1}H NMR spectrum of fraction G1 recorded at 30 °C in D_2O. The letters A, B, C, and D in the spectrum refer to corresponding residues in the structure (see Fig. 6).

spectrum. The H-1 signals at 5.18 and 5.20 ppm partly overlap, making it difficult to interpret the cross-peaks from these signals. The cross-peaks from these anomeric protons to the H-5 part of the spectrum are, however, slightly shifted downfield compared to the cross-peaks to H-2. Therefore, the H-1 signal at 5.18 ppm was assigned to a terminal arabinosyl group bound (1 → 2) (residue B, Fig. 6), and the signal at 5.20 was assigned to an arabinosyl residue substituted through O-2 and O-5 with the anomeric carbon bound (1 → 5) (residue D, Fig. 6). Finally, the H-1 signal at 5.30 ppm was assigned as for the arabinosyl residue at 5.20, but with the anomeric carbon bound (1 → 2) (residue C, Fig. 6). There were also some small, broad, and partly unresolved peaks in the anomeric region, that could not be assigned. Despite this, the main anomeric peaks could be integrated with the help of symmetric Lorentzian curve-fitting. The result showed a relationship between residues A:B:C:D (designations of residues according to Fig. 6) of 20:8:10:13, which agrees reasonably well with the methylation analysis result of an A + B to C + D ratio of 1.32. Combined, the results of methylation analysis and NMR experiments indicate that the main structural features of the water-soluble polysaccharide fraction G1 are those of an arabinan fragment (Fig. 6). In Fig. 6, there is admittedly an A + B to C + D ratio of 1.5, but this can be explained by some of residue X also being 2,5-substituted arabinose. We could accordingly assume that of the 34.6% of 2,5-substituted arabinose found by methylation analysis approximately 30% are C or D residues and 4% X residues. That would give a ratio of A + B + C + D to other arabinose residues (X) of approximately 5:1 according to methylation analysis, which fits with the structural element proposed in Fig. 6.

The small, broad peaks in the anomeric region of 5.1–5.5 ppm in the ^{1}H NMR

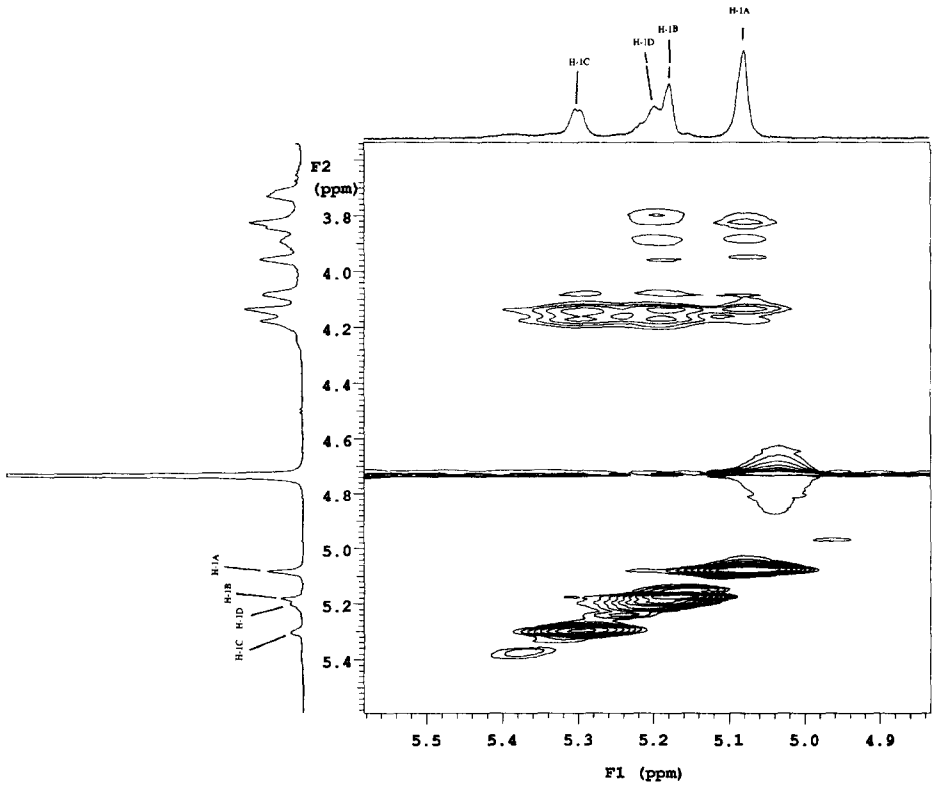

Fig. 5. 600-MHz $^{1}H,^{1}H$ NOESY spectrum of fraction G1 recorded at 30 °C in D_2O. The letters A, B, C, and D in the spectrum refer to corresponding residues in the structure (see Fig. 6).

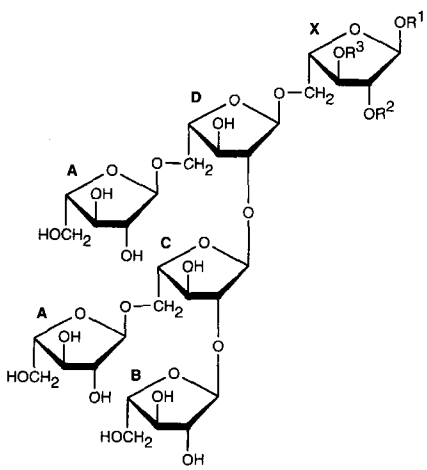

Fig. 6. Structural features of the gel filtration fraction G1 isolated from dehulled rapeseed meal. R^1, R^2, and R^3 illustrate that residue X can be bound in several different ways: to hydrogen, or to arabinose or galactose residues.

spectrum (Fig. 4) indicate the presence of small amounts of arabinose residues that are not part of the A–D structural features. There is also evidence of this from methylation analysis, where, for example, 4.0% of 5-substituted arabinose was detected. According to sugar analysis, 13.6% of the polysaccharide consists of galactose residues, and the results from methylation analysis (Table 2) show that 3,6-substituted and 3-substituted galactosyl residues predominate. However, it was not possible to assign any of the peaks as belonging to galactose in either of the NMR spectra. This was probably due to the relatively minor amount of galactose residues and because these residues were probably linked in different ways, the amounts of each type of galactose residue became even smaller. There are therefore insufficient data to determine how the arabinan fragment, suggested here, might be linked to galactose residues and/or other arabinose residues in the polysaccharide. A reasonable suggestion drawn from the data discussed above, however, is that the residues A–D are linked via C-5 of an unspecified arabinose residue (X, Fig. 6) to other arabinosyl residues or 3-substituted and 3,6-substituted galactosyl residues, illustrated by the different R-groups linked to X in Fig. 6.

To conclude, we have described a highly branched water-soluble polysaccharide, of which the main structural feature is an arabinan fragment. It is an unusual structure among arabinans in that it does not contain (1 → 5)-linked arabinose to any greater extent. Instead terminal arabinosyl groups and residues substituted at both C-2 and C-5 predominate. The presence of 3,6-substituted and 3-substituted galactosyl residues in fraction G1 suggests similarities to type II arabinogalactans [8,9,27], but 2,5-substituted arabinosyl residues are scarce in this type of polysaccharide. It is also unusual to find such a high ratio for Ara/Gal in an arabinogalactan. Further studies, perhaps involving enzymatic breakdown of the polysaccharide, are required to elucidate if and how the arabinan fragment described here is linked to the galactose residues.

Acknowledgements

Financial support was provided by the Swedish Council for Forestry and Agricultural Research, in collaboration with the Swedish Oil Extraction Ltd. and the Foundation of Swedish Oilseed Research/Association of Seed and Oilseed Growers.

References

[1] N.R. Swamy and P.V. Salimath, *Phytochemistry*, 30 (1991) 263–265.
[2] P. Capek, R. Toman, A. Kardošová, and J. Rosík, *Carbohydr. Res.*, 117 (1983) 133–140.
[3] J.-P. Joseleau, G. Chambat, and M. Lanvers, *Carbohydr. Res.*, 122 (1983) 107–113.
[4] B.J.H. Stevens and R.R. Selvendran, *Phytochemistry*, 19 (1980) 559–561.
[5] J.C. Villetaz, R. Amado, and H. Neukom, *Carbohydr. Polym.*, 1 (1981) 101–105.
[6] S.C. Churms, E.H. Merrifield, A.M. Stephen, D.R. Walwyn, A. Polson, K.J. van der Merwe, H.S.C. Spies, and N. Costa, *Carbohydr. Res.*, 113 (1983) 339–344.
[7] J.-P. Joseleau, G. Chambat, M. Vignon, and F. Barnoud, *Carbohydr. Res.*, 58 (1977) 165–175.
[8] A.E. Clarke, R.L. Anderson, and B.A. Stone, *Phytochemistry*, 18 (1979) 521–540.
[9] G.B. Fincher and B.A. Stone, *Annu. Rev. Plant Physiol.*, 34 (1983) 47–70.

[10] O. Larm, O. Theander, and P. Åman, *Acta Chem. Scand., Ser B.*, 30 (1976) 627–630.
[11] O. Larm, O. Theander, and P. Åman, *Acta Chem. Scand., Ser B.*, 29 (1975) 1011–1014.
[12] O. Theander and P. Åman, *Swed. J. Agric Res.*, 7 (1977) 69–77.
[13] I.R. Siddiqui and P.J. Wood, *Carbohydr. Res.*, 36 (1974) 35–44.
[14] I.R. Siddiqui and P.J. Wood, *Carbohydr. Res.*, 24 (1972) 1–9.
[15] I.R. Siddiqui and P.J. Wood, *J. Sci. Food Agric.*, 28 (1977) 530–538.
[16] I. Eriksson, E. Westerlund, and P. Åman, *J. Sci. Food Agric.*, 66 (1994) 233–240.
[17] O. Theander, P. Åman, E. Westerlund, R. Andersson, and D. Pettersson, *AOAC method 994.13, Official Methods of Analysis*, 16th ed., 1995, 1st supplement.
[18] F. Sosulski and R. Zadernowski, *J. Am. Oil Chem. Soc.*, 58 (1981) 96–98.
[19] E. Westerlund, R. Andersson, and P. Åman, *Carbohydr. Polym.*, 20 (1993) 115–123.
[20] P.J. Harris, R.J. Henry, A.B. Blakeney, and B.A. Stone, *Carbohydr. Res.*, 127 (1984) 59–73.
[21] P.-E. Jansson, L. Kenne, H. Liedgren, B. Lindberg, and J. Lönngren, *Chem. Commun. Univ. Stockholm*, (1976) 1–75.
[22] E.T. Oakley, D.F. Magin, G.H. Bokelman, and W.S. Ryan, *J. Carbohydr. Chem.*, 4 (1985) 53–65.
[23] D.P. Sweet, R.H. Shapiro, and P. Albersheim, *Carbohydr. Res.*, 40 (1975) 217–225.
[24] C. Bjergegaard, S.K. Jensen, and H. Sørensen, in D.I. McGregor (Ed.), *Proceedings GCIRC Eighth International Rapeseed Congress, Saskatoon, Saskatchewan, Canada, Vol. 2*, 1992, pp 448–453.
[25] B.A. Slominski, W. Guenter, and L.D. Campbell, *J. Agric. Food Chem.*, 41 (1993) 2304–2308.
[26] P.A.J. Gorin and M. Mazurek, *Carbohydr. Res.*, 48 (1976) 171–186; P.A.J. Gorin, *Adv. Carbohydr. Chem. Biochem.*, 38 (1981) 13–104.
[27] M. O'Neill, P. Albersheim, and A. Darvill, in P.M. Dey (Ed.), *Methods in Plant Biochemistry: Vol. 2, Carbohydrates*, Academic, London, 1990, pp 415–441.