ACTA CHEMICA SCANDINAVICA 15 (1961) 78–86

Studies on Arabogalactans

V. Barry Degradation of the Arabogalactans from Western Larch. A Kinetic Study of the Mild Acid Hydrolysis of Arabogalactan A

HANS O. BOUVENG

Träkemiska avdelningen, Svenska Träforskningsinstitutet, Stockholm Ö, Sweden

Arabogalactan A and B from Western Larch heartwood were subjected to periodate oxidation and then Barry degradation. Hydrolysis of degraded and methylated arabogalactan B gave 2,4-di-*O*-methyl-D-galactose, 2,4,6-tri-*O*-methyl-D-galactose and 2,3,4,6-tetra-*O*-methyl-D-galactose as main products together with smaller amounts of 2-*O*-methyl-D-galactose, 4-*O*-methyl-D-galactose, 2,6-di-*O*-methyl-D-galactose, 2,3,4-tri-*O*-methyl-D-galactose, 2,5-di-*O*-methyl-L-arabinose and 2,3,5-tri-*O*-methyl-L-arabinose.

The kinetics of the hydrolysis of arabogalactan A in 0.005 N hydrochloric acid were studied. The velocity constants for the hydrolysis of the L-arabinofuranose and 3-*O*-β-L-arabinopyranosyl-L-arabinofuranose residues were estimated as 7.4×10^{-4} and 2.9×10^{-4}, respectively, (min, Brigg's logarithms); that for the easily-hydrolysed galactose residues was not less than 1.0×10^{-4}.

Previous papers of this series have described the fractionation of the arabogalactans from Western larch (*Larix occidentalis* Nutt.) into two components, A and B, and an investigation of these by the methylation technique [1–3]. The complexity of polysaccharide A has been demonstrated [2]. In the present investigation the procedure developed by Barry [4] for the degradation of periodate-oxidised polysaccharides has been applied to the polysaccharides for the elucidation of further structural details.

Arabogalactan B consumed 0.96 mole of oxidant on periodate oxidation with the liberation of 0.44 mole of formic acid per mole of anhydrosugar. This is equivalent to an oxidation of 44 molar % of the residues in the polysaccharide as terminal and 6-substituted galactose and terminal, pyranosidic arabinose and of 8 % as terminal furanosidic arabinose, which agrees fairly well with methylation data (41.0 and 7.6 molar %, respectively) [3].

The oxidised polysaccharide was degraded by treatment with phenyl hydrazine and acetic acid as devised by Barry [4] to give a polysaccharide fraction together with glyoxal *bis*-phenylhydrazone, glyceroosazone and L-ara-

binoosazone. The lowmolecular weight fragments were identified by chromatography on dimethyl sulphoxide-impregnated paper [5]. Neither galactoosazone nor any oligosaccharide osazone could be detected. The degraded polysaccharide contained 10.9 molar % of arabinose. A calculation from methylation data of the arabinose content of the non-oxidisable residues in the original polysaccharide gave a value of 16.3 %. Thus about one third of the arabinose was split off as osazone during the degradation. This could be explained by direct hydrolysis of arabinofuranose residues during the degradation but the virtual absence of arabinoosazone among the products formed on degradation of oxidised arabogalactan A indicates however that the hydrolysis, if any, of furanosidic linkages is very minute under the conditions used. The results show rather that about one third of the 3-substituted arabinofuranose residues in polysaccharide B are linked through the 6-position to galactose residues carrying no other substituents (I) while the remainder and also the 3-substituted arabinofuranose residues in polysaccharide A are linked to non-oxidisable galactose residues, for instance II.

$$\beta\text{-L-Ara}p\ 1\text{———}3\ \text{L-Ara}f\ 1\text{———}6\ \beta\text{-D-Gal}p\ 1\text{———} \qquad \text{I}$$

$$\beta\text{-L-Ara}p\ 1\text{———}3\ \text{L-Ara}f\ 1\text{———}6\ \beta\text{-D-Gal}p\ 1\text{———} \qquad \text{II}$$
$$\qquad\qquad\qquad\qquad\qquad\qquad\qquad\qquad\qquad\qquad\qquad\qquad\qquad\qquad\quad 3$$
$$\qquad\qquad\qquad\qquad\qquad\qquad\qquad\qquad\qquad\qquad\qquad\qquad\qquad\qquad\quad |$$

The degraded polysaccharide was subjected to a further periodate oxidation and subsequent Barry degradation. A polymeric material could be isolated from the reaction mixture and on partial hydrolysis this gave as main product a homologuous series of oligosaccharides made up from β-1→3-linked galactose. Only faint traces of arabinose and of the 1→6-linked galactobiose were found. This makes it probable that the backbone of the polysaccharide consists of β-1→3-linked galactose residues.

Methylation of the first degradation product and hydrolysis of the methylated material gave the different methyl ethers shown in Table 1. The methylation steps were accompanied by severe losses, probably due to the low solubility of the methylated product and thus the results may not be fully representative for the original polysaccharide.

The high proportion of 2,4,6-tri-*O*-methyl-D-galactose gives further evidence of a backbone in the polysaccharide made up of 1→3-linked galactopyranose residues. The finding that practically no 1→6-linkages remain on further oxidation and Barry degradation of the polysaccharide suggests that the 2,4-di-*O*-methyl-D-galactose corresponds to residues in the main backbone carrying short branches, mainly single D-galactopyranose residues, at the 6-position. The 4-*O*-methyl-D-galactose is probably an artifact.

Table 1. Methyl ethers from methylated and hydrolysed degraded arabogalactan B.

Methyl ether	Molar %	Methyl ether	Molar %
2-Methylgalactose	3.2	2,4,6-Trimethylgalactose	38.9
4-Methylgalactose	1.6	2,3,4,6-Tetramethylgalactose	21.6
2,4-Dimethylgalactose	18.5	2,5-Dimethylarabinose	1.7
2,6-Dimethylgalactose	4.8	2,3,5-Trimethylarabinose	6.5
2,3,4-Trimethylgalactose	3.3		

Table 2. Methyl ethers corresponding to non-oxidisable residues from methylated and hydrolysed arabogalactan B.

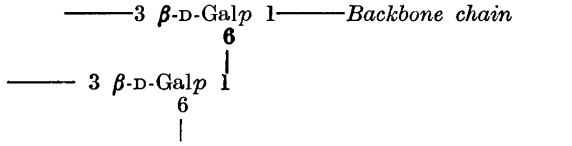
Methyl ether	Molar %	Methyl ether	Molar %
2-Methylgalactose	4.9	2,4,6-Trimethylgalactose	8.5
2,4-Dimethylgalactose	66.7	2,5-Dimethylarabinose	11.5
2,6-Dimethylgalactose	8.3		

The 2,3,5-tri-*O*-methyl-L-arabinose arises from 3-substituted arabinofuranose residues in undegraded arabogalactan B. The presence of 2,5-di-*O*-methyl-L-arabinose is in agreement with the previously assumed [3] occurrence of residues of type III in the original polysaccharide.

L-Ara(*p* or *f*) 1———3 L-Ara*f* 1———3 L-Ara*f* 1——— III

The methylation data given in Table 1 can profitably be compared with those obtained from the original polysaccharide. Table 2 shows the proportions of the methyl ethers [3] related to *non-oxidised* residues in arabogalactan B (the release of part of the arabinose as osazone is accounted for).

Comparison of Tables 1 and 2 shows that the main part of the terminal galactopyranose residues in the degraded polysaccharide originates from doubly substituted galactopyranose residues in the undegraded polysaccharide represented by the 2,4-di-*O*-methyl-D-galactose in Table 2. Heavily ramified elements like IV are thus not infrequent in arabogalactan B.

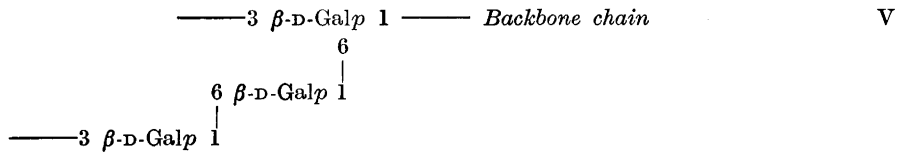


IV

As can be seen from Tables 1 and 2 there is an actual decrease of about 40 % in the residues corresponding to 2-*O*-methyl-D-galactose and 2,6-di-*O*-methyl-D-galactose. The remainder must be substituted by nonoxidisable (3-substituted) residues in the original polysaccharide.

As yet no unambiguous proof has been advanced that the 2-*O*- and 2,6-di-*O*-methyl-D-galactoses found in hydrolysates from methylated larch arabogalactans [1,3,6,7] have any structural significance. Partial hydrolysis would afford the corresponding 1,4-linked disaccharide but only in about the same amounts as would be expected to be formed by acid reversion of galactose [8]. A trial experiment was made, in which the hydrolysis was carried out in steps with intermediate withdrawal of low-molecular weight material in order to increase the yield of disaccharides. After fractionation of the hydrolysate by carbon column chromatography, one fraction was found by paper chromatography and electrophoresis in borate buffer to contain traces of a disaccharide with the same mobility as 4-*O*-β-D-galactopyranosyl-D-galactose [9]. However the large amounts of 3-*O*-β-D-galactopyranosyl-D-galactose accompanying it made any proper characterisation impossible.

Arabogalactan A consumed 1.01 mole of periodate with the liberation of 0.45 mole of formic acid per mole of anhydrohexose. This is equivalent to an oxidation of 45 molar % of the sugar residues in the polysaccharide as terminal and 6-substituted galactose and terminal, pyranosidic arabinose and of 11 % as terminal furanosidic arabinose. An estimate of these oxidisable residues from the methylation data [1] for comparison gave low values as the recovery of the corresponding methyl ethers was incomplete because of their slight volatility. Correction for these losses (*cf.* Ref.[3]) gave residues of 42 and 12 %, respectively, in fair agreement with the above values. No arabinoosazone or galactoosazone was observed among the degradation products formed on treatment with phenylhydrazine-acetic acid. The virtual absence of galactoosazone from the degradation products from the two polysaccharides excludes the presence of structural elements that contain 3-substituted galactose residues separated from the main chain by residues substituted only in the 6-position (V). Both degraded arabogalactans gave single, slightly oval spots with M_G values of 0.4—0.5 on electrophoresis on glass fibre sheets. Degraded polysaccharide A after mild hydrolysis unlike the original polysaccharide [2] gave one spot only on electrophoresis.



V

The results obtained in this series of investigations have shown few marked dissimilarities between the two arabogalactans from Western larch heartwood. Their molecular weights calculated from sedimentation constants differ considerably (4.3 and 1.4 for polysaccharide A and B, respectively). Polysaccharide A unlike B gives two distinct polymeric fragments when subjected to mild hydrolysis. The reason for this behaviour is not clearly understood.

The results of methylation and periodate oxidation studies show only slight differences in the proportions of different types of substituted galactose residues. The arrangement of the arabinose residues is, however, obviously different [1,3] but results of investigations of other larch arabogalactans [2,6,7] indicate that the arrangement as well as the amount of these residues can vary considerably in these polysaccharides. Polysaccharide A appears to be somewhat more branched than B. It also has a slightly higher proportion of residues that can form borate complexes. These residues include terminal and 6-substituted galactose and terminal pyranosidic arabinose, the total amount of which is equivalent to the amount of formic acid liberated on periodate oxidation of the polysaccharides. This slight difference does not, however, seem sufficiently large to account for the difference in rate of migration of the polysaccharides on electrophoresis in borate buffer (M_G values 0.76 and 0.63, respectively). Any comparison of the two polysaccharides in these respects is however open to criticism as long as the structural background of the splitting of polysaccharide A into the two polymeric fragments A I and A II [2] is not accounted for.

The results of this series of investigation and of investigations of similar arabogalactans (presumably of type A [2]) from European larch (*Larix decidua* Miller) [6,12] and tamarack (*L. laricina* (Du Roi) Koch) [1] make it possible to outline a general structure for these complicated polysaccharides. They contain a backbone made up mainly, or possibly exclusively, of β-1→3-linked D-galactopyranose residues. This backbone is ramified by substitution at the 6-positions and the low proportion of 2,4,6-tri-*O*-methyl-D-galactose found in the hydrolysates from the methylated polysaccharides indicates that few if any of the 6-positions in these backbone residues are unsubstituted. No regular pattern for the side-branching is discernible. Calculations from the data obtained from methylated Western larch arabogalactan B and from the products of periodate oxidation and Barry degradation, indicate that two thirds of the residues in this polysaccharide occur in the side-chains, that is, each side-chain contains an average of two residues. The non-terminal galactose seems largely to be 6-substituted and there are indications of residues in the side-chains of the B polysaccharide that are substituted in both the 3- and 6-positions. The arrangement of the arabinose is presumably variable. It occurs as single terminal furanosidic residues, as biosidic residues and to some extent as chains of three residues (III).

The structural significance of the 2-*O*- and 2,6-di-*O*-methyl-D-galactose found in the hydrolysates from methylated larch arabogalactans has not been unequivocally proved. Undermethylation or demethylation during hydrolysis might explain their appearance but this is contradicted by their virtual absence from the hydrolysate of an otherwise similar arabogalactan isolated from maple sap.[13]

Several investigations [6,10,11] have shown that significant amounts of D-galactose and 6-*O*-β-D-galactopyranosyl-D-galactose are liberated in addition to furanosidically linked L-arabinose and 3-*O*-β-L-arabinopyranosyl-L-arabinose when larch arabogalactans are subjected to acid hydrolysis under very mild conditions, for instance 0.01 N acid at 100°. This suggests that D-galactofuranosidic residues are present in the polysaccharide but careful investigations [3,6] have failed to give any positive evidence for the presence of such residues.

A semi-quantitative estimate was made of the hydrolysis constants for L-arabinose and 3-*O*-β-L-arabinopyranosyl-L-arabinose and for D-galactose together with 6-*O*-β-D-galactopyranosyl-D-galactose in the hydrolysis of arabogalactan A with 0.005 N hydrochloric acid at about 95°. Samples were withdrawn at suitable intervals and the amounts of the different saccharides were estimated by hypoiodite oxidation [14] after separation on thick filter paper. The results are shown in Table 3. The velocity constants are calculated assuming a monomolecular reaction, that is $kt = -\log (c_o - c)/c_o$, and extrapolated to time 0. When the hydrolysis had proceeded for 9 h, about half of the arabinose originally present had been split off and the k values had decreased up to 15 %. Methylation data gave the amount (c_o) of each saccharide in the starting material. However, the amount of galactose and 6-β-galactobiose originally accessible to hydrolysis under these conditions is not known and suitable values must be chosen arbitrarily. The first value (25 molar % of the original polysaccharide) assumes that the main part of the terminal galactose residues is hydrolysed at a uniform rate while the second value (10 %)

Table 3. Velocity constants (min. and Brigg's logarithms) for the hydrolysis of some saccharides from arabogalactan A in 0.005 N hydrochloric acid at 95°.

Saccharide	$k \times 10^4$
L-Arabinofuranose	7.4
3-*O*-β-L-Arabinopyranosyl-L-arabinofuranose	2.9
D-Galactopyranose + 6-*O*-β-D-galactopyranosyl-D-galactopyranose, 25 %	1.0
» , 10 %	2.5

corresponds to the minimum value given by the results from various experiments that have been made in this series of investigations on the mild hydrolysis of the polysaccharide.

As can be seen from Table 3 these galactopyranosidic residues are hydrolysed at almost the same rate as the arabinofuranosidic. The hydrolysis rates for pyranosides and furanosides normally differ [15] by a factor of about 10^3.

EXPERIMENTAL

All melting points are corrected. All evaporations were done under reduced pressure.

Chromatography. Papers: Whatman No. 1 and Schleicher and Schüll 602 hP. Solvents: Butanol, ethanol, water, 10:3:5, and benzene, dimethyl sulphoxide (DMSO), 20:1 (for chromatography on DMSO-impregnated paper [5]). Spray reagent: anisidine hydrochloride. The osazones required no spray reagent.

Electrophoresis. Papers: Whatman No. 1 and Schleicher and Schüll glass fibre sheets. Buffer: 0.1 M borate buffer of pH 10. Spray reagents: Anisidine hydrochloride and α-naphthol-sulphuric acid in butanol.

Periodate oxidation of arabogalactan B. Periodate oxidations were done in 0.125 M sodium periodate at room temperature. The oxidation of arabogalactan B was complete in 4 h, 0.96 mole of periodate being consumed and 0.44 mole of formic acid liberated. The overoxidation was 0.03 mole in 24 h.

Barry degradation of oxidised arabogalactan B. Arabogalactan B (10 g) was oxidised for 6 h and then ethylene glycol was added to destroy excess periodate. The solution was freed from iodate by precipitation with lead acetate and filtration. Excess lead ion was precipitated with sulphuric acid. The solution was filtered and neutralised and then phenylhydrazine (25 ml) and acetic acid (12 ml) were added. The yellow flocculent precipitate formed was recovered on a glass filter and thoroughly washed with water. It contained 11.57 % nitrogen. An oxidised polysaccharide-phenylhydrazine complex of the type described by Barry *et al.*[16] and with a composition in accord with the methylation data for arabogalactan B [3] would contain 11.68 % nitrogen.

The precipitate was dispersed in aqueous ethanol (400 ml), acetic acid (60 ml) and phenylhydrazine (30 ml) and refluxed for 4 h. The dark-coloured solution was concentrated and the glyoxal *bis*-phenylhydrazone which crystallised (m.p. and mixed m.p. 168—170°) was removed by filtration. Water was then added to the solution and it was exhaustively extracted, first with benzene and then with ether. The extracts were examined by chromatography on paper impregnated with DMSO [5]. As well as glyoxal *bis*-phenylhydrazone this also indicated the presence of glycerosazone and L-arabinoosazone. No galactoosazone was observed. The ether extract contained mainly phenylhydrazine acetate.

The extracted aqueous solution was concentrated and run into ethanol to give the degraded arabogalactan as a yellow precipitate. After purification by reprecipitation the polysaccharide was obtained as a pale yellow powder (3.94 g) containing 3.1 % nitrogen. It was later found that the nitrogen content could be greatly reduced by treatment of the polysaccharide with hot 20 % acetic acid for 20—60 min. Arabinose constituted 10.9 % of the reducing sugars in the polysaccharide.

Table 4. Fractionation of methylated and hydrolysed, degraded arabogalactan B.

Fract.	mmole (tot)	Sugars	%	mmole
24—32	0.096	2-Methylgalactose	66	0.063
		4-Methylgalactose	34	0.033
33—48	0.342	2,4-Dimethylgalactose		
49—60	0.120	2,4-Dimethylgalactose	21	0.025
		2,6-Dimethylgalactose	79	0.095
61—80	0.100	2,3,4-Trimethylgalactose	66	0.066
		2,5-Dimethylgalactose	34	0.034
81—143	0.775	2,4,6-Trimethylgalactose		
Residue	0.558	2,3,4,6-Tetramethylgalactose	77	0.430
		2,3,5-Trimethylarabinose	23	0.129

Parts of the extracts and the filtrate from the precipitated polysaccharide were taken to dryness and then dissolved in ethanol and treated with Amberlite IR 120 in the acid form. An oligosaccharide osazone would, according to Finan and O'Colla [17], be hydrolysed by this treatment to give a reducing saccharide with one residue less. No reducing saccharides were detected on chromatography of the supernatant liquid and thus no larger fragments could have been split off as osazones during the degradation.

Methylation of degraded arabogalactan B. The degraded polysaccharide (3.0 g) was methylated first with dimethyl sulphate in aqueous 30 % sodium hydroxide, then twice with methyl iodide and barium oxide in dimethylformamide (DMFA) [2,18] and finally twice with methyl iodide and silver oxide in DMFA. The final product (0.85 g) was difficult to free from impurities and it was not possible to obtain a reliable methoxyl analysis. No unmethylated monomer was detected on chromatography of a hydrolysed sample.

Hydrolysis of methylated, degraded arabogalactan B and fractionation of the hydrolysate. The methylated polysaccharide (0.6 g) was first treated with hot formic acid (50 ml) for 1 h. After evaporation of the formic acid it was hydrolysed completely by using 0.5 N sulphuric at 100° for 16 h. This procedure has been found to cause very little degradation and demethylation. The hydrolysate was fractionated on a carbon-Celite column (3.5 × 46 cm) using 5 l 6→35 % aqueous ethanol as eluant. Mixed fractions were subfractionated on thick filter paper and the amount of reducing sugar in the various fractions was estimated by hypoiodite oxidation [14]. The results of the fractionations are summarised in Tables 1 and 4.

*Characterisation of the methyl ethers. 2-O-Methyl-*D*-galactose, 4-O-methyl-*D*-galactose* and *2,6-di-O-methyl-*D*-galactose* were indistinguishable from authentic specimens on paper chromatography and on paper electrophoresis.

*2,5-Di-O-methyl-*L*-arabinose* and *2,3,4-tri-O-methyl-*D*-galactose* were indistinguishable from authentic specimens on normal paper chromatography and on chromatography on paper impregnated with DMSO.

*2,4-Di-O-methyl-*D*-galactose.* The ether was recrystallised from ethyl acetate-methyl ethyl ketone, m.p. 100°; the aniline derivative had m.p. 216—218°, undepressed on admixture with authentic 2,4-di-*O*-methyl-D-galactosyl-*N*-phenylamine.

*2,4,6-Tri-O-methyl-*D*-galactose.* The ether was recrystallised from ethanol-*iso*propyl ether, m.p. 101—104°; the aniline derivative had m.p. 176—178°, undepressed on admixture with authentic 2,4,6-tri-*O*-methyl-D-galactosyl-*N*-phenylamine.

*2,3,4,6-Tetra-O-methyl-*D*-galactose.* The aniline derivative had m.p. 199—200°, undepressed on admixture with authentic 2,3,4,6-tetra-*O*-methyl-D-galactosyl-*N*-phenylamine.

*2,3,5-Tri-O-methyl-*L*-arabinose.* The ether was converted to the amide of the corresponding acid. It had m.p. 138—140° after recrystallisation from ethyl acetate, in good agreement with values previously reported [19].

Periodate oxidation and Barry degradation of arabogalactan A. Arabogalactan A on oxidation with periodate consumed 1.01 mole of oxidant with the liberation of 0.45 mole of formic acid per mole anhydrohexose. The oxidised polysaccharide was degraded in the same manner as arabogalactan B to yield a polysaccharide material containing 8.1 %

arabinose (arabinose content calculated from methylation data 6.2 %). Neither L-arabinoosazone nor D-galactoosazone was observed among the degradation products.

Miscellaneous experiments on degraded arabogalactans A and B. Both polysaccharides yielded arabinose only on hydrolysis in 0.01 N hydrochloric at 100° for 2 h. They travelled as single spots on electrophoresis in borate buffer both before and after mild hydrolysis.

When subjected to graded hydrolysis (0.1 N hydrochloric acid), both afforded 3-*O*- and 6-*O*-β-D-galactopyranosyl-D-galactose but the proportion of the latter was less than in case of the original polysaccharides.

Degraded arabogalactan B was subjected to a second periodate oxidation followed by Barry degradation. As the polysaccharide gave a rather dark solution, no estimate of the consumption of oxidant could be made. A polysaccharide fraction could be isolated as a brownish powder and this on hydrolysis for 2 h in 0.1 N hydrochloric acid at 100° gave the following saccharides as main products (the fractions R_M/n^{20} are given in brackets): D-galactose (0.36), 3-*O*-β-D-galactopyranosyl-D-galactose (0.35), a trisaccharide (0.35), a tetrasaccharide (0.33) and higher oligosaccharides. The similarity of the R_M/n fractions indicates that the polymeric material consisted mainly of straight chains of β-1→3-linked D-galactopyranose residues. Only faint traces of arabinose and 6-*O*-β-D-galactopyranosyl-D-galactose appeared on the chromatogram.

Partial hydrolysis of arabogalactan A. Arabogalactan A (4.0 g = 25.6 mmole of anhydrosugar) was hydrolysed in 0.005 N hydrochloric acid (200 ml) at about 95°. Samples of suitable size (2—4 ml) were withdrawn at intervals and run into ethanol to precipitate polymeric material which was removed by filtering. After reprecipitation of the solids and filtering, the oligosaccharides in the combined filtrates were separated on prewashed Schleicher and Schüll 602 hP papers. The amounts of L-arabinose, D-galactose and 6-*O*-β-D-galactopyranosyl-D-galactose were estimated by hypoiodite oxidation directly after elution from the papers. As the 3-*O*-β-L-arabinopyranosyl-L-arabinose might not be stable at the pH used for the hypoiodite oxidation [21], the eluate containing it was taken to dryness and dissolved in hot ethanol. Any hemicellulosic material from the paper was removed by filtering the hot solution. The solvent was evaporated and the disaccharide was hydrolysed in 0.5 N sulphuric acid. The solution was neutralised with sodium bicarbonate and the amount of arabinose present was estimated by hypoiodite oxidation. The results of the hydrolysis are given in Table 5.

Calculation of the hydrolysis constants. The velocity constants for the hydrolysis of L-arabinose, 3-β-arabinobiose and of D-galactose together with 6-β-galactobiose were calculated from the curves constructed from the values given in Table 5. Only the values from the first nine hours should be taken into consideration as the secondary hydrolysis of the disaccharides taking place during this short period can probably be disregarded. Methylation data [1] gave the following proportions in molar % of hydrolysable residues (c_0) in the starting material: arabinofuranose 12 % = 3.07 mmole; 3-β-arabinobiose 3 % = 0.77 mmole; D-galactose + 6-β-galactobiose 25 % = 6.40 mmole (assuming that the main part of the terminal galactose and galactobiose residues was hydrolysed at an uniform rate) and 10 % = 2.56 mmole (assuming that only part of the residues was sensitive to hydrolysis under the conditions used). The results of the calculation are summarised in Table 3.

Table 5. Saccharides formed on hydrolysis of 4.0 g of arabogalactan A in 0.005 N hydrochloric acid at 95° (mmole).

Time, h	L-arabinose	D-galactose	3-*O*-β-L-arabinopyranosyl-L-arabinose	6-*O*-β-D-galactopyranosyl-D-galactose
0.5	0.165			
1	0.267	0.073	0.027	
2	0.636	0.150	0.056	0.036
4	1.04	0.242	0.097	0.048
6	1.37	0.368	0.165	0.092
8.5	1.66	0.452	0.216	0.102
23.5	2.70	1.13	0.284	0.194
30	2.88	1.43	0.270	0.238

The author is greatly indebted to Professor Bengt Lindberg for his keen interest in this work and to Miss Anita Myhrman for her skilful assistance.

REFERENCES

1. Bouveng, H. O. and Lindberg, B. *Acta Chem. Scand.* **12** (1958) 1977.
2. Bouveng, H. O. *Acta Chem. Scand.* **13** (1959) 1869.
3. Bouveng, H. O. *Acta Chem. Scand.* **13** (1959) 1877.
4. Barry, V. C. *Nature* **152** (1943) 538.
5. Wickberg, B. *Acta Chem. Scand.* **12** (1958) 615.
6. Aspinall, G. O., Hirst, E. L. and Ramstad, E. *J. Chem. Soc.* **1958** 593.
7. Adams, G. A. *Can. J. Chem.* **38** (1960) 280.
8. Thompson, A., Anno, K., Wolfrom, M. L. and Inatome, M. *J. Am. Chem. Soc.* **76** (1954) 1309.
9. Bouveng, H. O. and Meier, H. *Acta Chem. Scand.* **13** (1959) 1884.
10. Jones, J. K. N. *J. Chem. Soc.* **1953** 1672.
11. Bouveng, H. O. and Lindberg, B. *Acta Chem. Scand.* **10** (1958) 1515.
12. Aspinall, G. O., Cairncross, J. M. and Nicholson, A. *Proc. Chem. Soc.* **1959** 270.
13. Bishop, C. T. *Private communication.*
14. Hirst, E. L., Hough, L. and Jones, J. K. N. *J. Chem. Soc.* **1949** 57.
15. Pigman, W. W. and Goepp, R. M. *Chemistry of the Carbohydrates*, New York 1948, p. 206.
16. Barry, V. C. and Mitchell, P. W. D. *J. Chem. Soc.* **1953** 3631.
17. Finan, P. A. and O'Colla, P. S. *Chem. & Ind. London* **1955** 1387.
18. Kuhn, R., Baer, H. H. and Seeliger, A. M. *Ann.* **611** (1958) 236.
19. Maher, G. G. *Advances in Carbohydrate Chem.* **10** (1955) 263.
20. French, D. and Wild, G. M. *J. Am. Chem. Soc.* **75** (1953) 2612.
21. Whistler, R. L. and BeMiller, J. N. *Advances in Carbohydrate Chem.* **13** (1958) 289.

Recived September 9, 1960.