

544. Further Oligosaccharides from Carboxyl-reduced Gum Arabic

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EVIDENCE concerning the location of L-rhamnopyranose residues in *Acacia senegal* gum (gum arabic) was obtained by the characterisation of 4-*O*- α -L-rhamnopyranosyl-D-glucose as one of the products of depolymerisation of the carboxyl-reduced gum.¹ It was observed that higher oligosaccharides containing rhamnose residues had also been formed. One oligosaccharide fraction, which gave rhamnose as one hydrolysis product, has now been separated chromatographically into three components. The first component was characterised as 3-*O*- β -D-galactopyranosyl-D-galactose by the isolation of the crystalline disaccharide. The second component was assigned the structure, *O*- α -L-rhamnopyranosyl-(1 \rightarrow 4)-*O*- β -D-glucopyranosyl-(1 \rightarrow 6)-D-galactose, on the basis of the following observations. Hydrolysis of the trisaccharide gave rhamnose, glucose, and galactose, whilst hydrolysis of the derived glycitol gave rhamnose, glucose, and galactitol, showing the presence of a galactose reducing group. Under mild conditions of hydrolysis rhamnose was released before the other monosaccharides indicating that this sugar residue occupied a terminal position in the oligosaccharide. A sample of the trisaccharide was methylated and hydrolysis of the methylated derivative furnished 2,3,4-tri-*O*-methyl-L-rhamnose, 2,3,6-tri-*O*-methyl-D-glucose, and 2,3,4-tri-*O*-methyl-D-galactose. Although the methylated derivative was inadvertently hydrolysed before methylation was quite complete with the consequent formation of di-*O*-methyl derivatives of glucose and galactose (but not of rhamnose), in addition to the above sugars, the absence of other trimethyl ethers leaves little doubt as to the structurally significant methylated sugars. Since gum arabic furnishes 6-*O*- β -D-glucopyranosyluronic acid-D-galactose as a partial hydrolysis product,² and since the D-glucopyranosyl residues in the carboxyl-reduced gum are formed from D-glucopyranosyluronic acid residues, it follows that the neutral sugar residues will have the β -configuration. The observed optical rotation ($[\alpha]_D +10^\circ$) of the trisaccharide is consistent with this assignment if it is also assumed that the L-rhamnopyranose residue has the α -configuration as in the disaccharide, 4-*O*- α -L-rhamnopyranosyl-D-glucose.¹ The third oligosaccharide component was tentatively assigned the structure, *O*- β -galactopyranosyl-(1 \rightarrow 3)-*O*- β -D-galactopyranosyl-(1 \rightarrow 3)-D-galactose, since partial hydrolysis gave galactose and only one disaccharide which had the chromatographic mobility of the 1,3-linked disaccharide. The galactobiose, which had been isolated previously on partial hydrolysis of the unmodified gum,³ and the galactotriose clearly had their origin in the main chain of the polysaccharide which is known to be composed largely, if not exclusively, of sugar residues linked in this manner.⁴

Experimental.—The general experimental conditions were as described by Aspinall *et al.*¹

Fractionation of oligosaccharide fraction 4c. Fraction 4c (0.5 g.) from the acetolysis of carboxyl-reduced gum arabic¹ was separated chromatographically on filter sheets using solvent A to give fractions A, B, and C.

Fraction A. The sugar (91 mg.), $R_{\text{galactose}}$ 0.51 in solvent A and $[\alpha]_D +58^\circ$ (c 0.48), gave galactose only on hydrolysis. The sugar was recrystallised from acetone–water to give 3-*O*- β -D-galactopyranosyl-D-galactose monohydrate, which was identified by m. p. and mixed m. p. 158° , and by X-ray powder photograph.

Fraction B. The sugar (170 mg.), $R_{\text{galactose}}$ 0.35 in solvent A and $[\alpha]_D +10^\circ$ (c 0.43), gave rhamnose, glucose, and galactose on hydrolysis. Reduction of the oligosaccharide (10 mg.) in water (1 ml.) with sodium borohydride (10 mg.) gave the glycitol, hydrolysis of which afforded rhamnose, glucose, and galactitol [detected by chromatography in butan-2-one–acetic acid–water (9:1:1, saturated with boric acid)]. Partial hydrolysis of the oligosaccharide with 0.1N-sulphuric acid for 1 hr. on the boiling-water bath resulted in the release of rhamnose and the formation of a new oligosaccharide ($R_{\text{galactose}}$ 0.45). Oligosaccharide B (165 mg.) was methylated successively with methyl sulphate and sodium hydroxide, and methyl iodide and

¹ G. O. Aspinall, A. J. Charlson, E. L. Hirst, and R. Young, *J.*, 1963, 1696.

² S. W. Challinor, W. N. Haworth, and E. L. Hirst, *J.*, 1931, 259.

³ J. Jackson and F. Smith, *J.*, 1940, 79.

⁴ F. Smith and R. Montgomery, "Chemistry of Plant Gums and Mucilages," Reinhold Publ. Corp., New York, 1959.

silver oxide to give the methylated derivative (140 mg.). The whole of the methylated derivative was inadvertently hydrolysed, and paper chromatography of the hydrolysate and gas chromatography of the derived methyl glycosides indicated the presence of 2,3,4-tri-*O*-methyl-rhamnose, 2,3,6-tri-*O*-methylglucose, and 2,3,4-tri-*O*-methylgalactose, together with smaller amounts of di-*O*-methylhexoses. The mixture (130 mg.) of methylated sugars was separated on cellulose (60 × 2 cm.) with light petroleum (b. p. 100–120°)–butan-1-ol (7 : 3, later 1 : 1), saturated with water, as eluant to give five fractions. Fraction 1 (40 mg.), $[\alpha]_D +23^\circ$ (*c* 0.4), was characterised as 2,3,4-tri-*O*-methyl-L-rhamnose by conversion into the aniline derivative, m. p. and mixed m. p. 114°. Fraction 2 (16 mg.), $[\alpha]_D +72^\circ$ (*c* 0.16), was characterised as 2,3,6-tri-*O*-methyl-D-glucose by conversion into the 1,4-di-*p*-nitrobenzoate, m. p. and mixed m. p. 193°. Fraction 3 (12 mg.), $[\alpha]_D +85^\circ$ (*c* 0.12), contained a mixture of 2,3,6-tri-*O*-methylglucose and 2,3,4-tri-*O*-methylgalactose. Fraction 4 (21 mg.), $[\alpha]_D +118^\circ$ (*c* 0.21), was characterised as 2,3,4-tri-*O*-methyl-D-galactose by conversion into the aniline derivative, m. p. and mixed m. p. 154°. Fraction 5 (34 mg.) gave only one spot on paper chromatography, but gas chromatography of the derived methyl glycosides showed the presence of several components and demethylation gave glucose and galactose.

Fraction C. The sugar (89 mg.), $R_{\text{galactose}} 0.17$ in solvent *A* and $[\alpha]_D +56^\circ$ (*c* 0.89) (Haq and Adams⁵ quote $[\alpha]_D +51^\circ$ for the 1,3-linked galactotriose), gave galactose only on hydrolysis. Partial acid hydrolysis gave only one disaccharide with the chromatographic mobility of 3-*O*-β-D-galactopyranosyl-D-galactose.

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⁵ S. Haq and G. A. Adams, *Canad. J. Chem.*, 1961, **39**, 1563.