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NOTE

Methylation of *Acacia auriculiformis* gum polysaccharide

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Polysaccharide has been extracted from *Acacia auriculiformis* gum with water as L-arabinose and D-galactose in 1 : 4 molar ratio. Methylation of gum afforded methyl sugars as 2,3,4,6-tetra-O-methyl-D-galactose; 2,3-di-O-methyl-L-arabinose; 2,3,4-tri-O-methyl-D-galactose; 2,4-di-O-methyl-D-galactose and 2,3,4-tri-O-methyl-D-glucuronic acid in 1 : 1 : 1 : 2 : 1 molar ratio.

Acacia auriculiformis plant[1] (Mimoseae) occurs in Northern India, and the gum contain a water soluble polysaccharide. Present manuscript deals with methylation studies alongwith a proposed gum polysaccharide structure.

Paper chromatography was carried out by descending technique[2] on Whatman No. 1 and 3 mm paper with upper phase of the following solvent system (v/v): (S_1) *n*-butanol, ethanol, water (4 : 1 : 5)[3], (S_2) *n*-butanol, acetic acid, water (4 : 1 : 5)[3] and (R_1) *p*-anisidine phosphate[4] used as spray reagent for the detection of methyl sugars.

Methylation of gum polyaccharide: Polysaccharide (15 g) was methylated by Hakomari's method[5] 3 times with water (40 mL), dimethyl sulphate (175 mL) and sodium hydroxide (30%, 150 mL) in atmospheric nitrogen at 5–10°C giving partly methylated product (6.50 g, —OCH_3, 38%). It was remethylated 5 times by Purdie's reagent[6] with methanol (30 mL), methyl iodide (30 mL) and silver oxide (12 g), which gave fully methylated product (5.024 g, —OCH_3, 42.58%), $[\alpha]_D^{27}$–39.8° (H_2O).

Hydrolysis of methylated gum polysaccharide: Methyl sugar (5 g) was refluxed with methanolic hydrogen chloride (4%, 15 mL), then evaporated off to dryness and saponified with barium hydroxide (0.03 N, 10 mL) at 60°C for 3 h. Excess $Ba(OH)_2$ was removed by CO_2 in the solution. Saponified product after

being worked up give an ether soluble portion (A) (yield 2.002 g and an ether insoluble portion (B) (yield 0.3756 g).

Identification of ether soluble portion (A): It (2.002 g) was hydrolysed with HCl (1 N, 40 mL) on water-bath (10 h). Hydrolysate was cooled, neutralised ($BaCO_3$) and evaporated to syrup. It was resolved into 4 components on Whatman No. 3 mm paper (S_1) and paper strips corresponding to individual sugars were eluted with water[7].

I: 2,4-*di-O-methyl-D-galactorse*: Syrup (0.5992 g) gave single spot on paper chromatography (S_1), R_f (0.40), $[\alpha]_D^{27}$ +81.2°C (H_2O). Found: —OCH_3, 29.8%. Derivative was prepared by refluxing syrup (0.3 g) with aniline (0.162 g) and ethanol (5 mL) giving crystals of 2,4-di-O-methyl-N-phenyl-D-galactosyl amine, m.p. 208°C.[8]

II: 2,3,4-*tri-O-methyl-D-galactose*: Syrup (0.351 g) having $[\alpha]_D^{27}$ +114° (H_2O) showed single spot on paper chromatography (R_f 0.62 in S_1), Found: —OCH_3, 39.7%, calcd. for $C_9H_{18}O_6$, —OCH_3, 41.9%. Derivative was prepared as 2,3,4-tri-O-methyl-N-phenyl-D-galactosyl amine, m.p. 163°C.[8]

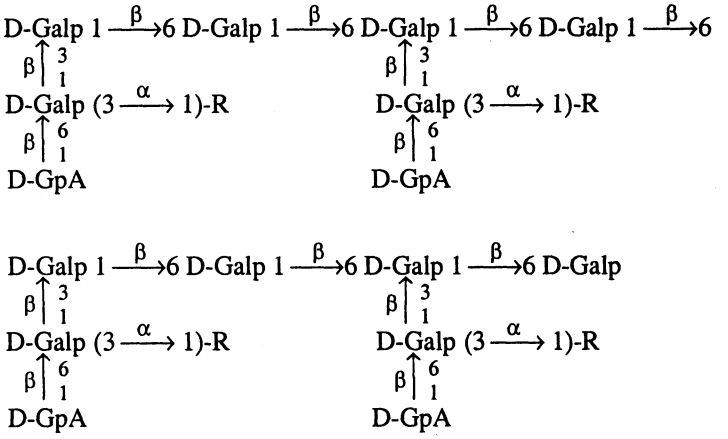
III: 2,3-*di-o-methyl-L-arabinose*: Syrup (0.2814 g) gave single spot (R_f 0.63 in S_1) on paper chromatography, $[\alpha]_D^{27}$ +98° (H_2O). Found: —OCH_3, 34.9%, calcd, for $C_7H_{14}O_5$, 34.8%. Sugar was converted into 2,3-di-O-methyl-N-phenyl-L-arabinose amine derivative, m.p. 138°C.[9].

IV: 2,3,4,6-*tetra-O-methyl-D-galactose*: Syrup (0.310 g) had R_f, 0.86 (S_1), $[\alpha]_D^{27}$ +112° (H_2O). Found: —OCH_3, 51.99%; calcd. for $C_{10}H_{20}O_6$, 52.5%. It was converted into 2,3,4,6-tetra-O-methyl-N-phenyl-D-galactosyl amine, m.p. 190–191°C (192°C).[10]

Identification of ether insoluble portion (B): Syrup (0.3756 g) on paper chromatography in S_2 used (R_1) as spray reagent to reveal single spot (R_f, 0.82) Found: —OCH_3, 38.4%, calcd. for 2,3,4-tri-O-methyl-D-glucuronic acid, $C_9H_{16}O_6$, 39.4%. Derivative was prepared by usual manner as 2,3,4-tri-O-melthyl-γ-D-glucopyranoside uronomide, m.p. 183°C.[8]

Acacia auriculiformis gum polysaccharide was methylated by Hakomari and Purdie's method. Methyl sugars were identified as 2,3,4-tetra-O-methyl-D-galactose, 2,3-di-O-methyl-L-arabinose, 2,3,4-tri-O-methyl-D-galactose, 2,4-di-O-methyl-D-galactose and 2,3,4-tri-O-methyl-D-glucuronic acid in 1 : 1 : 1 : 2 : 1 molar ratio. 2,3,4,6-tetra-O-methyl-D-galactose suggest that side chain R is terminated by D-galactopyranose moiety. Side chain R may be regarded as composed of L-arabofuranose unit joined to a D-galactopyranose unit (5→1)-α-type linkage, accordingly the R = L-Araf (5→1)-D-Galp. The uronic acid moiety is justified an acidic sugar, 2,3,4-tri-O-methyl-D-glucuronic acid. It may be pointed out here that in gum structure, L-fucose is not included because it was obtained in traces. Hydrolysis of methylated gum did not furnish any methylated L-fucose unit.

The tentative structure (Fig. 1) suggested for *A. auriculiformis* gum polysaccharide clearly indicates that gum is highly branched in nature and contains (1→6)-β, (1→3)-β and (1→5)-α-type linkages.



where: Galp = Galactopyranose; GpA = Glucopyranosyl uronic acid

R = L-Araf (5 $\xrightarrow{\alpha}$ 1)-D-Galp

Fig.1 Tentative structure of *Acacia auriculiformis* gum polysaccharide

REFERENCES

1. Y.R. Chadha, The Wealth of India: Raw Materials, CSIR, New Delhi, vol. 1-A, p. 23 (1985).
2. S.M. Partridge, *Nature* (*London*), **158**, 270 (1946).
3. S.M. Partridge and R.G. Westall, *Biochem. J.*, **42**, 238 (1948).
4. S. Mukherjee and H.C. Srivastava, *Nature* (*London*), **169**, 330 (1952).
5. S. Hakomari, *J. Biochem.* (*Tokyo*), **55**, 205 (1964).
6. T. Purdie and J.C. Irvine, *J. Chem. Soc.*, **83**, 1021 (1903).
7. C.E. Dent, *Biochem. J.*, **41**, 240 (1947).
8. S. Bose and K.C. Gupta, *Indian J. Chem.*, **2**, 156 (1964).
9. E.L. Hirst and J.K.N. Jones, *J. Chem. Soc.*, 120 (1948).
10. F. Smith, *J. Chem. Soc.*, 1035 (1940).

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