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Note

Structural studies of the O-specific side-chains of the lipopolysaccharide from *Yersinia enterocolitica* Ye 128

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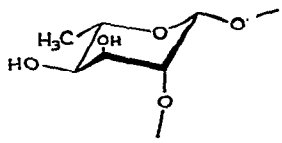
The presence of the unusual sugar 6-deoxy-L-altrose as a component of the lipopolysaccharide (LPS) from a strain of *Yersinia enterocolitica* has been reported[1]. Sugar analyses of LPS from strains of this organism indicate that this sugar is present in LPS from different serotypes[2,3]. We now report studies of the LPS from *Y. enterocolitica* Ye 128 (Daniels 924) belonging to serotype 2 of Winblad[4] or serogroup IB of Knapp and Thal[5].

The polysaccharide (PS) was prepared from the LPS by acid hydrolysis under mild conditions. An acid hydrolysate of the PS contained a sugar which was chromatographically indistinguishable from 6-deoxy-D-altrose[6], together with traces of glucose, heptose, and another minor component, tentatively identified as rhamnose. The major component was isolated as a chromatographically pure syrup. Its ^{1}H-n.m.r. spectrum and that given by authentic 6-deoxy-D-altrose were superposable. The natural sugar has $[\alpha]_{578}$ −14° (*c* 2, water), compared to +16° for the D sugar[7], demonstrating that the former has the L configuration.

Methylation analysis of the PS gave a single, major component, identified from the mass spectrum of its alditol acetate as 6-deoxy-3,4-di-*O*-methyl-L-altrose. Consequently, the sugar residues in the PS are pyranosidic and linked through the 2-position.

The PS had $[\alpha]_{578}$ +72° (*c* 1, water), indicating that the 6-deoxy-L-altropyranosyl residues have the β configuration. In agreement with this conclusion, the signal for the anomeric proton in the ^{1}H-n.m.r. spectrum appeared at δ 5.16 ($J_{1,2}$ low), compared to the value δ 5.09 ($J_{1,2}$ 1.3 Hz) reported for β-D-altropyranose[8]. The presence of only six strong signals in the ^{13}C-n.m.r. spectrum confirmed that the major component of the PS was the O-antigenic chain, having a simple structure. The signal for C-1 appeared at 101.3 p.p.m. with a $J_{^{13}C,H}$ value of 164 Hz, demonstrating that H-1 is axial[9]. The combined evidence therefore demonstrates that the

O-antigen is a homopolymer composed of (1→2)-linked 6-deoxy-β-L-altropyranosyl residues, and that these residues are in the $^1C_4$ conformation, as in **1**.



1

EXPERIMENTAL

General methods. — Concentrations were performed under reduced pressure at bath temperatures not exceeding 40°. G.l.c. was performed with a Perkin–Elmer model 990 instrument fitted with flame-ionisation detectors. Separations were performed on 3% of OV-225 on Gas Chrom W (100–200 mesh) in glass columns (180 × 0.15 cm). G.l.c.–m.s. was performed on a Varian Mat 311 instrument fitted with an OV-225 column. For n.m.r. spectra, a Jeol FX 100 instrument was used. The spectra were obtained for solutions in D_2O at 85°, using external tetramethylsilane (^{13}C-n.m.r.) or internal sodium 1,1,2,2,3,3-hexadeuterio-4,4-dimethyl-4-silapentane-1-sulfonate (^{1}H-n.m.r.) as references. Undecoupled spectra were obtained by the gated decoupling technique with a sampling time of 0.4 sec and a pulse repetition time of 1 sec. Optical rotations were determined with a Perkin–Elmer 241 instrument. Methylation was performed by the Hakomori method[10] with sodium methylsulfinylmethanide–methyl iodide in dimethyl sulfoxide. Hydrolysis and analysis of the products were performed as previously described[11].

Identification of sugar component. — Acid hydrolysis of the polysaccharide gave one major component, identified by g.l.c.–m.s. of its alditol acetate as a 6-deoxyhexose. This sugar was separated by p.c. on Whatman No. 1 paper, using 1-butanol–pyridine–water (6:4:3) as irrigant.

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