

BASIC STRUCTURAL FEATURES OF A LIPOPOLYSACCHARIDE FROM THE *COXIELLA BURNETII* STRAIN NINE MILE IN THE VIRULENT PHASE I

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Lipopolysaccharide (LPS) located on the surface of the *Coxiella burnetii* virulent phase I cells has been considered (1, 2) to be a major determinant of virulence but despite of this fact little is known (3) about its primary structure. In our recent paper (4) the chemical composition and tentative distribution of constituent sugars in this LPS have been reported. Moreover, it has been found that the chemical structure of *C. burnetii* LPS in phase I seems to represent a significant departure from the structures described for enteric LPSs.

Highly purified LPS (5 mg) was solubilized in dry dimethyl sulfoxide (2 ml) and methylated with methyl iodide (1.5 ml) in the presence of methylsulphonyl carbanion (5). The solution was then poured into water (5 ml), dialyzed in distilled water for 48 hr, and evaporated. The sirupy residue was dissolved in methyl iodide (5 ml), silver oxide (20 mg) was added (6), and the mixture was stirred and boiled under reflux for 24 hr. The procedure was repeated once. The fully methylated LPS (3 mg) was hydrolyzed with 2 mol/l trifluoroacetic acid (1.5 ml) in a sealed tube at 105 °C for 16 hr and the methylated saccharides were conventionally converted into the corresponding alditol acetates and analyzed (7) by GLC - mass spectrometry. The latter technique was performed with a JMS-D 100 (Jeol) spectrometer, using a column (200 cm × 0.3 cm) packed with 0.124-0.147 mm. Supelcoport coated with 3 % of SP 2340. The inlet helium pressure was 101.3 kPa at a programmed temperature range of 150 °C (4 min) to 220 °C at 2 °C min<sup>-1</sup>. The spectra were determined at 23 eV. The following methylated sugars could be identified as the corresponding alditol acetates: 2, 3, 4-tri-0-methylvrenose, 2, 3, 5-tri-0-methyl-3-C-/methoxy/-dihydro-hydroxystreptose, 2, 3, 4, 6-tetra-0-methyl-D-mannose, 2, 3, 6-tri-0-methyl-D-mannose, 4, 6, 7-and 2, 6, 7-tri-0-methyl-D-glycero-D-mannoheptose, 3-0-methylrhamnose, 4, 6-di-0-methyl-N-/methyl/acetylglucosamine, and 3, 4-di-0-methyl-N-/methyl/acetylglucosamine.

In conclusion, the results suggest that the following kinds of sugar units might be present in the LPS under investigation:

	↓	↓
	4	4
Vir-/1→	→3/-D-Hep-/1→	→2/-Rhp-/1→
Strep-/1→	→3/-D-Hep-/1→	→3/-D-GlcNAc-/1→
D-Manp-/1→	2	→6/-D-GlcNAc-/1→
	↑	

Vir, virenopyranose (6-deoxy-3-C-methylgulopyranose); Strep, dihydro-hydroxystreptofuranose (3-C-/hydroxymethyl/-lyxofuranose); D-Hep, D-glycero-D-mannoheptose

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