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# Structure of the capsular polysaccharide of *Acinetobacter baumannii* 1053 having the KL91 capsule biosynthesis gene locus



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# ABSTRACT

Acinetobacter baumannii 1053 is the type strain for the maintenance of specific bacteriophage AP22, which infects a fairly broad range of *A. baumannii* strains circulating in Russian clinics and hospitals. A capsular polysaccharide (CPS) was isolated from cells of strain 1053 and studied by sugar analysis along with 1D and 2D <sup>1</sup>H and <sup>13</sup>C NMR spectroscopy. The following structure of the linear trisaccharide repeating unit was established:

 $\rightarrow$ 4)- $\beta$ -D-ManpNAcA-(1 $\rightarrow$ 4)- $\beta$ -D-ManpNAcA-(1 $\rightarrow$ 3)- $\alpha$ -D-FucpNAc-(1 $\rightarrow$ 

where ManNAcA and FucNAc indicate 2-acetamido-2-deoxymannuronic acid and 2-acetamido-2,6dideoxygalactose, respectively. A polysaccharide having the same repeating unit but a shorter chain was isolated by the phenol–water extraction of bacterial cells. Sequencing of the CPS biosynthesis gene locus showed that *A. baumannii* 1053 belongs to a new group designated KL91. The gene functions assigned putatively by a comparison with available databases were in agreement with the CPS structure established.

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Recently, *A. baumannii* has become one of the most widespread agents causing health-care associated infections. Treatment of these infections is complicated owing to the ability of the bacteria to acquire and to accumulate various antibiotic resistance mechanisms.<sup>1</sup> *A. baumannii* lacks a long-chain lipopolysaccharide with an O-polysaccharide chain<sup>2</sup> but has a capsular polysaccharide (CPS) that forms a thick layer around the bacterial cell. The CPS protects *A. baumannii* from the action of immune system components and is considered as an important virulence factor.<sup>3</sup> A number of *A. baumannii* CPS structures have been established to date (some under the wrong name of O-antigen or O-specific polysaccharide) (Refs. 4–9 and refs. cited in Ref. 4) but more remain to be elucidated.

Bacteriophages that are able to infect *A. baumannii* represent a promising alternative to antibiotics to control this pathogen. From

http://dx.doi.org/10.1016/j.carres.2014.11.013 0008-6215/© 2014 Elsevier Ltd. All rights reserved. 130 *A. baumannii* isolates collected by us from clinics and hospitals in Russia in 2005–2010, 89 isolates (~68%) were sensitive to Myoviridae bacteriophage AP22,<sup>10</sup> making it suitable for development of anti-acinetobacter preparations. To infect the host bacteriophages have to disrupt the CPS layer, and CPS is the primary receptor for the phage AP22 structural depolymerase/ adsorption protein (data will be published elsewhere). In this work, we studied structure and genetics of the CPS of *A. baumannii* 1053, which is the type strain for the phage AP22 maintenance.

CPS was isolated from cells of *A. baumannii* 1053 by extraction with phosphate-buffered saline containing EDTA. Sugar analysis of CPS by GLC of the acetylated alditols revealed 2-amino-2,6-dide-oxygalactose (fucosamine, FucN). GLC analysis of the acetylated (*S*)-2-octyl glycosides showed that FucN has the D configuration.

The <sup>13</sup>C NMR spectrum of the CPS (Fig. 1) showed signals for three anomeric carbons at  $\delta$  98.9–100.9, CH<sup>3</sup>–C groups of one 6-deoxyhexose (C-6 of FucN) at  $\delta$  16.6 and three *N*-acetyl groups at  $\delta$  23.2–23.3, three nitrogen-bearing carbons at  $\delta$  48.8–54.7, other

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