anchored LAM and LM will be presented to illustrate the principles and strengths of the FAB-MS sequencing strategies.

S19.7 Studies on Glycine as a Component of Bacterial Lipopolysaccharides

A. Gamian, E. Katzenellenbogen, M. Mieszała, E. Romanowska and A. Czarny Institute of Immunology and Experimental Therapy, Polish Academy of Sciences, Wroclaw, Poland.

The aminoacyl analysis of endotoxic lipopolysaccharides (LPS) isolated from several bacteria revealed the essential amounts of glycine, among the inherent LPS components. This prompts for the detailed investigation in view of the search for common epitopes suitable for construction of antimicrobial vaccine with broad specificity. The glycine, possibly unifying the LPS structures, might create the common epitope. Significant amounts of glycine was detected in lipopolysaccharides isolated from over 30 strains of Shigella, Escherichia coli, Salmonella, Hafnia and Citrobacter. When the O-specific polysaccharide, lipid A and core fractions of Shigella flexneri and sonnei LPSs were subjected to aminoacyl analysis, glycine as a single aminoacid was found only in core oligosaccharide. The quantity of the aminoacid was not stoichiometric in core oligosaccharides. Molar ratio of glycine ranged from 0.2 to 0.6 per 3 heptoses. Using the HPLC the oligosaccharide enriched in glycine was isolated. Mild acid treatment (0.05 M HC1, 15 min, 100°C) released free glycine. The aminoacid could be also partially cleaved (in 50%) by pronase digestion. Growing E. coli C600 or Sh. sonnei phase II in the presence of radioactive ¹⁴C-Gly, the labelling of their lipopolysaccharide cores was achieved. The experiments indicate that glycine is covalently bound component of core oligosaccharide in bacterial lipopolysaccharides.

S19.8

Studies on the O-Antigen of Pseudomonas Syringae PV. Tomato

G. M. Zdorovenko, L. P. Solyanik, L. M. Yakovleva and Y. A. Knirel

Department of Biochemistry, Institute of Microbiology and Virology of the Ukrainian Academy of Sciences, Kiev, Ukraine.

P. syringae pv. tomato is the agent of tomato bacterial diseases. O-antigen from the wet microbial cells was extracted with 0.85% NaCl. The preparation contained all common components of O-antigenic macromolecule, as carbohydrates, lipid A, protein and was active in the tests with homologous O-antiserum against microbial cells. It induced water-soaking lesions on tomato leaves and tuberous tissue on cabbage. O-antigen was not toxic to HeLa line cells (carcinoma of cervix of the uterus) but its in vivo injection into mice previously inoculated with ascitic Erlich's carcinoma decreased the longevity of the animals for about 27%. Structural parts of O-antigenic molecule, as lipid A, core oligosaccharides, O-polysaccharide (O-PS) were obtained and examined separately. Compositions of lipid A and core fractions were common for typical Pseudomonas. Basing on ¹H and ¹³C NMR spectroscopy data, its computer analysis, etc, the structure of the repeating unit O-PS was elucidated:

³L-Rha
$$\frac{1,2}{\alpha}$$
L-Rha $\frac{1,3}{\alpha}$ L-Rha $\frac{1}{\alpha}$
D-Fuc³NAc

S19.9

Chemical Structure of Three O-Specific Polysaccharides of Proteus Penneri Lipopolysaccharides

Z. Sidorczyk¹, A. Świerzko², E. V. Vinogradov³, Y. A. Knirel³

¹Institute of Microbiology and Immunology, University of Ľódź, Ľódź, Poland; ²Centre of Microbiology and Virology, Polish Academy of Sciences, Ľódź, Poland; ³Institute of Organic Chemistry, Russian Academy of Sciences, Moscow, Russia.

Proteus penneri is a novel species distinguished in genus Proteus on the basis of DNA hybridization experiments and physiological properties. High antigenic heterogeneity of Proteus strains is associated with composition and structure of O-specific polysaccharide chains of outer membrane lipopolysaccharides.

Three O-specific polysaccharides of P. penneri (8, 52 and 62) are structurally elucidated with the help of 1D and 2D NMR spectroscopy (including 1D TOCSY and NOE, 2D COSY, 1 step and 2 step COSYRCT, ¹³C/¹H COSY and ROESY) and chemical degradations (O-deacetylation, O-dephosphorylation, partial hydrolysis, solvolysis and Smith degradation). These antigens contain D-Glc, D-Gal, D-GlcN, DGalN, L-FucN, D-GalA as main sugar constituents. 2-amino-2-deoxy sugars carry N-acetyl group, the other non-sugar components found are O-acetyl group, residue of lactic acid at position 3 of D-GlcNAc and 2-aminoethyl group attached via phosphate group at position 6 of D-GlcNAc. They are built up of linear or branched tetra-, penta- and hexasaccharide repeating units, respectively. Each of them contain from two up to five charged groups — acidic or/and basic functions. Three serologically distinguishable strains were found to possess O-antigen with the unique structure. The immunodominant role of some lateral sugar residues or charged substituents was established.

S19.10

Chemical Structure and Serological Specificity of Proteus Mirabilis 028 O-Specific Polysaccharide and Lipopolysaccharide from a Transient Form of Proteus Mirabilis R14/1959

J. Radziejewska-Lebrecht¹, E. V. Vinogradov², B. Bartodziejska, Y. A. Knirel², W. Kaca¹, H. Mayer³, A. Chernyak² and A. Rózalski¹

¹Institute of Microbiology and Immunology, University of Łódź, Łódź, Poland; ²Institute of Organic Chemistry, Russian Academy of Sciences, Moscow, Russia; ³Max-Planck Institut für Immunbiologie, Freiburg, Germany.

Proteus mirabilis is Gram-negative bacterial pathogen which causes UTI. Serological studies revealed the close relatedness of LPS from P. mirabilis 028 and R14/1959. The strain R14/1959 reminiscent Salmonella T-forms. On the basis of structural analysis it was concluded the following structure of investigated antigens: