

## ESCHERICHIA COLI P2B POLYSACCHARIDE AS A LIGAND FOR COMPLEMENT LECTIN PATHWAY ACTIVATING FICOLIN-2 AND 3

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Ficolin-2 and ficolin-3 are important factors of innate immunity. They are able to participate in elimination of pathogens through opsonisation of their cells (contributing to phagocytosis) or complement activation *via* the lectin pathway (thanks to co-operation with serine proteases of the MASP family), leading to the direct cell lysis. They may form heterocomplexes, called ficolin-23. While ficolin-3 specificity is restricted to very few microorganisms, ficolin-2 recognises numerous bacteria, fungi, parasites, and viruses. Among its ligands, capsular polysaccharides, lipoteichoic acids, mycobacterial lipoarabinomannan and antigen 85 complex, *Streptococcus pneumoniae* pneumolysin,  $1,3-\beta$ -glucans, and hemagglutinin of influenza A virus are mentioned. Moreover, artificial ligands like acetylated albumin (Ac-BSA) or N-acetylglucosamine pentamer (GN5-DPPE) were previously used to detect active ficolin-2 molecules (although no ficolin-2-dependent complement activation by Ac-BSA was observed) [1,2].

Here we report interaction of ficolin-2 and ficolin-3 with *Escherichia coli* P2b (serotype O15) bacteria, isolated from blood of preterm neonate, born with signs of intrauterine infection. Both ficolin-2 and ficolin-3 (as well as murine ficolin A, but not human ficolin-1 or mannose-binding lectin) were found to recognise high molecular mass fraction of supernatant obtained after ultracentrifugation of *E. coli* P2b lipopolysaccharide (LPS). Upon recognition of that fraction by ficolin-2/ficolin-3, MASP-1 enzyme became activated leading to complement lectin pathway activation.

Structural analyses of composition of the supernatant indicated the presence of O15 LPS, enterobacterial common antigen (ECA<sub>PG</sub> and ECA<sub>CYCLIC</sub>), and capsular polysaccharide (CPS, antigen K) built up of the  $[\rightarrow 3)$ - $\alpha$ -D-GlcpNAc-(1 $\rightarrow$ P]<sub>n</sub>. Immunoblotting and ELISA analyses indicated CPS as a functional ligand for ficolin-2 and 3, and excluded O15 LPS and ECA. Identified *E. coli* P2b CPS represents previously described K51 antigen reported as  $[\rightarrow 3)$ - $\alpha$ -D-GlcpNAc-(1 $\rightarrow$ P]<sub>n</sub> with O-acetyl groups at 4 and 6 position [3], however contrary to the published structure, identified CPS was devoid of O-acetyl groups.

## **References:**

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