

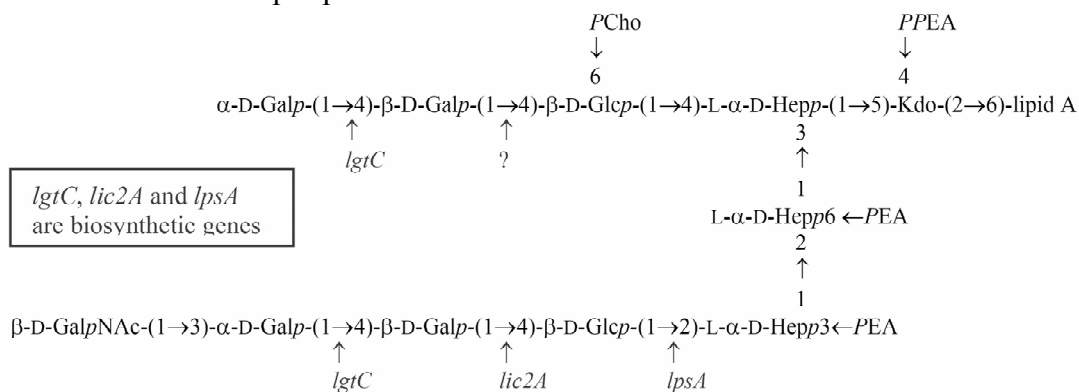
STRUCTURE AND FUNCTIONAL GENOMICS OF LIPOPOLYSACCHARIDE EXPRESSION IN NONTYPEABLE *HAEMOPHILUS INFLUENZAE*

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Common structural motifs of *Haemophilus influenzae* lipopolysaccharide (LPS) are globotetraose (β -D-GalpNAc-(1 \rightarrow 3)- α -D-Galp-(1 \rightarrow 4)- β -D-Galp-(1 \rightarrow 4)- β -D-Glcp) and its truncated versions globoside (α -D-Galp-(1 \rightarrow 4)- β -D-Galp-(1 \rightarrow 4)- β -D-Glcp) and lactose (β -D-Galp-(1 \rightarrow 4)- β -D-Glcp) linked to the terminal heptose (HepIII) of the triheptosyl inner-core moiety. We report here on structural studies of LPS from nontypeable *H. influenzae* strain 1124 expressing these motifs linked to both the proximal heptose (HepI) and HepIII at the same time [1]. This novel finding was obtained by structural studies of LPS using NMR techniques and electrospray ionization mass spectrometry (ESI-MS) on *O*-deacylated LPS and core oligosaccharide material (OS) as well as ESI-MS_n on permethylated dephosphorylated OS. The use of defined mutants allowed us to confirm structures unambiguously and understand better the biosynthesis of each of the globotetraose units. We found that *lgtC* is involved in the expression of α -D-Galp-(1 \rightarrow 4)- β -D-Galp in both extensions, whereas *lic2A* directs only the expression of β -D-Galp-(1 \rightarrow 4)- β -D-Glcp when linked to HepIII. The LPS of NTHi strain 1124 contained sialylated glycoforms that were identified by CE-ESI-MS/MS. A common sialylated structure in *H. influenzae* LPS is sialyllactose linked to HepIII. This structure exists in strain 1124. However, results on the *lpsA* mutant indicate that sialyllactose extends from HepI as well, a molecular environment for sialyllactose in *H. influenzae* that has not been reported previously. We are currently investigating the biosynthetic pathways leading to the expression of the above mentioned epitopes.



References

1. Yildirim, H. H.; Li, J.; Richards, J. C.; Hood, D. W.; Moxon, E. R.; Schweda, E. K.H.; *Biochemistry*, **2005**, *44*, 5207-5224.