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Funding Source: NSF-REU Program in Biological Sciences & Biochemistry

Autoaggregation and adhesion by nontypeable Haemophilus influenzae strains with mutations in the autotransporter Lav

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Nontypeable Haemophilus influenzae (NTHi) is a nonencapsulated gram-negative coccobacillus that colonizes the upper respiratory tract of most healthy adults.

Pathogenic NTHi cause otitis media, sinusitis, bronchitis, pneumonia, and complicate COPD and cystic fibrosis. Many NTHi from clinical isolates (but not commensal NTHi) encode Lav, homologous to bacterial autotransporter proteins. Lav is a phase-variable outer membrane protein whose expression is ON or OFF depending on the number of GCAA repeats following the initiating ATG codon. Preliminary experiments suggested that Lav improves adherence to lung tissue culture cells, but is not a primary adhesin. As some adhesins affect bacterial autoaggregation, a component of biofilm formation, we compared mutants with a knockout mutation in Lav to wild-type and phase-locked ON strains and found that null mutants caused unusually rapid autoaggregation of the null mutant relative to strains expressing Lav. However, the rapidly aggregating null mutant was seen to form filaments, leading us to suspect that its construction affected expression of neighboring genes (e.g., tmk) essential for DNA synthesis. Starting with the phase-locked ON construct, I constructed new mutants deleted for Lav that were fully isogenic with positive control strains. We will repeat autoaggregation and cell adherence assays using these more appropriate knockout mutants.