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“Investigating the Role of LtuA during the Developmental Cycle of *Chlamydia trachomatis*”

Introduction: *Chlamydia trachomatis*, an obligate intracellular Gram-negative bacterium, is the leading infectious cause of blindness in developing countries, as well as the most prevalent sexually transmitted bacterium. There is currently no vaccine available. *C. trachomatis* characteristically alternates between two forms in its developmental cycle: infectious non-replicative elementary bodies (EBs) and replicative non-infectious reticulate bodies (RBs). EBs transition to RBs in the early stage, RBs multiply in the middle stage, and RBs differentiate into EBs in the late stage. The growth of this pathogen correlates with a unique pattern of gene expression at each of the stages. However, little is known about how *C. trachomatis* controls its developmentally regulated gene expression, due partially to a historic lack of genetic tools to manipulate genes in *Chlamydia*. This study is focused on chlamydial *ltuA*. Although *ltuA* has been shown to be expressed in the late stage at the transcriptional level, the function of LtuA remains elusive.

Methods: In order to determine the functional role of *ltuA* during the chlamydial developmental cycle, we used a recently developed genetic transformation technique. For this, *ltuA* was cloned into an *E. coli-Chlamydia* shuttle plasmid so that *ltuA* expression could be controlled by a tetracycline inducible promoter. PCR, restriction enzyme digestion, agarose gel electrophoresis, and DNA sequencing were used to determine the identity of the construct containing *ltuA*.

Results: We successfully constructed the plasmid carrying *ltuA*. This expression vector will be transformed into *C. trachomatis* cells using our established approach. We will assess whether the conditional expression of LtuA can be achieved by adding tetracycline at the optimal concentration in the transformed bacteria and whether LtuA expression may affect the bacterial growth phenotype.

Conclusions. This study uses advanced genetic techniques to understand the functional role of LtuA. It is expected that the effects of the gene product of *ltuA* can be determined by analysis of the growth patterns and temporary gene expression profile in *C. trachomatis*.