

## ***luxS* Expression Influences *Escherichia coli* Biofilm Macrostructure**

KELLY J. PITTMAN, Chandan Robbins, Bryan A. Stubblefield and Eric S.

Gilbert

Biology

Faculty Sponsor: Eric Gilbert

*Escherichia coli* resides in the mammalian gut as a member of a microbial biofilm, or a community of microorganisms attached to a surface. One of the benefits for *E. coli* associated with living in a biofilm is enhanced resistance to antibiotics. The enhanced resistance is in part derived from the three-dimensional structure of the biofilm, which creates a diffusional barrier that shelters the inhabitant microbes and also a redox gradient that may impair the inhibitory action of antibiotics. Thus the ability to alter biofilm structure chemotherapeutically could benefit antibiotic therapies targeting pathogenic *E. coli*. A factor affecting biofilm formation for some microorganisms is quorum sensing, or cell-cell signaling. Quorum sensing in *Escherichia coli* involves the interspecies signaling molecule autoinducer-2 (AI-2), synthesized by AI-2 synthase and regulated by *luxS*, and regulates types including virulence factor production and flagella mediated motility. We hypothesized that *luxS* could potentially influence *E. coli* biofilm structure. To test this hypothesis, we compared biofilm formation by a *luxS* deletion mutant of *E. coli* W3110, which cannot produce AI-2, and a *luxS*-overexpressing variant that produces AI-2 at high concentrations. Biofilms were cultivated in parallel plate flow cells using an assay that accelerated their formation over a 5 hour period and were subsequently imaged by confocal laser scanning microscopy for quantitative analysis of biofilm structure. Digital image analysis using the program COMSTAT quantified several parameters characterizing biofilm structure of each strain, and determined significant phenotypic differences. Notably, the biomass, average thickness, and substratum coverage of the overexpressed variant was at least 1.5 times as much as the deletion mutant. Conversely, the roughness of the biofilm was almost 4 times greater in the deletion mutant than in the overexpressed strain. These results support the hypothesis, and indicate that *luxS* could potentially serve as a target for drugs that potentiate antibiotic activity against *E. coli* in biofilms.