In vivo Microarray Screening to Identify Uropathogenic *Escherichia coli* (UPEC) Genes Involved in Early Colonization and Infection in Murine Urinary Tract Infections

Kelly M. Winterberg*[†], Fredrick R. Blattner[‡], Rodney A. Welch[††], William S. Reznikoff [†]
[‡] Department of Genetics, [††] Department of Medical Microbiology and Immunology, [†] Department of Biochemistry, University of Wisconsin, Madison, WI, 53706 USA

Uropathogenic *Escherichia coli* (UPEC) is the cause of over 70% of all urinary tract infections (UTIs). To identify genes involved in early colonization and infection of the bladder, we have implemented a screening technique that combines transposon mutagenesis and oligonucleotide microarray analysis. Insertions are generated using a transposon containing divergent T7 promoters and a kanamycin resistance cassette. A set of 2,068 Tn*5* UPEC mutants (strain CFT073) have been generated and stored in 96-well format. As an attenuation control, a *fimH* knockout mutant was constructed and placed twice in each 96-well dish. Biotin-labeled RNA transcripts, generated from input (inoculum) and output (recovered) pools, are hybridized to custom designed oligonucleotide microarrays containing probes for both the top and bottom strands of the entire CFT073 genome, to determine insertion locations (gene identification) for individual mutants. Pools of 50 mutants are currently being screened in the murine UTI model where the bladders are harvested 6 hours post-inoculation. We will present a summary of these results and discuss mutant confirmation and follow-up strategies. This work will elucidate CFT073 mutants that are attenuated *in vivo*, and ultimately identify genes that are required for UPEC to colonize and/or survive in the bladder. Once identified, these genes may serve as potential therapeutic targets for treating bladder infections.