

KENDALL SOUDER, MICHAEL WHABY, JENNIFER HICKMAN, Department of Natural Sciences and Mathematics, West Liberty University, West Liberty, WV, DONALD PRIMERANO, JAMES DENVIR, Genomics and Bioinformatics Core Facility, Marshall University, Huntington, WV, and DEANNA M. SCHMITT. Department of Natural Sciences and Mathematics, West Liberty University, West Liberty, WV. Role of FTL\_1306 (*dipA*) in *Francisella tularensis* susceptibility to resazomycins.

The CDC classifies *Francisella tularensis* as a Category A bioterrorism agent. Due to the risk of potential release of antibiotic-resistant *F. tularensis* strains, new therapeutics against *F. tularensis* must be developed. Resazomycins are resazurin (Rz)-based compounds that exhibit antimicrobial activity against *F. tularensis* and other gram-negative bacteria. The action of resazomycins is not well understood, but potential targets of the antibiotic were identified in a high throughput screen for Rz-resistant isolates. The *dipA* (FTL\_1306) gene was identified as mutated in half of the 48 Rz-resistant (RZR) strains sequenced. To further investigate the role of *dipA* in Rz susceptibility, we introduced a wild-type copy of *dipA* into select RZR isolates (RZR1, 5, 43, and 46) that contain *dipA* mutations. The *dipA* gene was amplified by PCR from wild-type *F. tularensis* and cloned into the *F. tularensis* shuttle vector pABST to generate a construct (pABST-*dipA*) in which *dipA* will be constitutively expressed under control of the *groEL* promoter. The pABST-*dipA* plasmid was mobilized into each of the selected RZR isolates by electroporation. The resulting RZR *dipA*-complemented strains were cultivated on chocolate agar containing 10xMIC of Rz to determine their susceptibility to resazomycins. Preliminary results suggest that expression of wild-type *dipA* in the RZR mutants did not restore sensitivity to Rz indicating *dipA* may not play a role in Rz susceptibility. Further investigation is needed to fully elucidate the contribution of *dipA* to the bactericidal action of resazomycins.