EMMA J. BEATTY, EMILY YOUNG, CLAIRE KELLY, RORI SCHRIEBER, JORDAN GIBSON, KENDALL SOUDER, JUSTIN RICE, NICOLE GARRISON, and DEANNA M. SCHMITT, Department of Biomedical Sciences, West Liberty University, West Liberty, WV 26074, and RYAN J. PERCIFIELD, Department of Biology, West Virginia University, Morgantown, WV 26506, and DONALD A. PRIMERANO, Department of Biomedical Sciences, Joan C. Edwards School of Medicine, Marshall University, Huntington, WV 25755.

Role of LpnA in *Francisella tularensis* Susceptibility to Resazurin.

Francisella tularensis is defined by the Centers for Disease Control and Prevention as a Category A bioterrorism agent due to its low infectious dose, high mortality rate, and ease of aerosolization. Given there is no licensed tularemia vaccine in the United States and the possible development and release of antibiotic-resistant *F. tularensis* strains, there is an urgent need for new treatments against this bacterium. We determined the phenoxazine dye resazurin (Rz) exhibits antimicrobial activity against *F. tularensis* and other gram-negative bacteria. The mode of action of this compound is not understood, but potential targets of Rz were identified in a high throughput screen for resistant isolates. Ninety-three percent of the Rz-resistant (Rzr) isolates sequenced contained mutations within the coding regions of FTL_0421 (*lpnA*), FTL_0895, and FTL_1504 (*katG*). To confirm mutation of *lpnA* was contributing to Rz resistance, we introduced a wild-type copy of *lpnA* into Rzr1 and then assessed the susceptibility of the resulting complemented strain (Rzr1/pABST-*lpnA*). The minimum inhibitory concentration (MIC) of Rzr1 and Rzr1/pABST-*lpnA* were similar, therefore, we wanted to confirm we were restoring expression of LpnA in the complemented strain via Western blot analysis. As expected, LpnA was expressed in wild-type LVS but not in Rzr1 due to the genetic mutation. In the Rzr1 strain transformed with pABST-*lpnA*, we still did not observe expression of LpnA suggesting our complementation strategy was ineffective. In the future, we plan to take an alternative approach to investigate the role of LpnA in resazurin resistance by creating a *lpnA* deletion mutant.