J Bacteriol Parasitol 2018, Volume: 9
DOI: 10.4172/2155-9597-C1-044

6th International Congress on
Bacteriology & Infectious Diseases
May 21-22, 2018 | New York, USA

Identification of a novel gene in ROD9 island of *Salmonella enteritidis* involved in the alteration of virulence-associated genes expression

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Salmonella enterica subsp. I serovar *enteritidis* (*S. Enteritidis*), one of the causative agents for non-typhoidal gastrointestinal diseases in humans is an intracellular bacterium and mechanism for its invasion into host cells is critical to cause infection. The virulence of the pathogen is explained by the expression of genes located on its pathogenicity islands, mostly encoded under SPI-1 and SPI-2. However, *S. typhimurium* SL1344, despite sharing ~98% of its genome with S. *Enteritidis* P125109, lacks few regions of differences (ROD) that are hypothesized to impart virulence potential to S. *Enteritidis*. In this study, we created different mutants in the ROD9 island of S. *Enteritidis*, also referred as SPI-19 and identified a novel locus, SEN1005, encoding a hypothetical protein that is involved in its pathogenesis. ΔSEN1005 displayed significantly reduced entry into cultured epithelial cells as well as uptake by macrophages and failed to cause acute colitis in C57BL/6 mice at day three post-infection (p.i.). Additionally, the global transcriptome analysis revealed a highly repressed SPI-1 and other down-regulated genes responsible for flagellar assembly, chemotaxis and motility in the mutant which correlated with decreased invasion and abated inflammation as compared to the wild-type. Therefore, our findings revealed that ΔSEN1005 was attenuated in vitro as well as *in vivo* and we propose this hypothetical protein to play a role in altering the expression of genes involved in *Salmonella* virulence.

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