

## Investigating the role of the toxin-encoding bacteriophage and host background on the expression of Stx2 in *Escherichia coli* O157:H7

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The foodborne pathogen *Escherichia coli* O157:H7 expresses one or more Shiga toxins (Stx), which are responsible for the serious clinical manifestations of infections in humans. Stx2, which is associated with highly pathogenic strains, is encoded within the genome of a lambdoid prophage and expressed when the prophage is induced. It has also been hypothesized that the abundance of phage and susceptible commensal *E. coli* in the intestinal tract also impacts Stx2 accumulation. In this study, we quantified phage and Stx2 produced by 13 *E. coli* O157:H7 strains and observed dramatic strain-to-strain variability. In order to address how genetic components of these toxin-encoding phage were responsible for the levels of phage and toxin production, we generated phage genome sequences from 9 strains within our collection. By transducing the *stx2*-encoding phage from *E. coli* strain EDL933 into non-toxin producing *E. coli* and *S. flexneri* strains, we determined that the host background affects phage and toxin production. We also observed that *stx2*-encoding phage from different *E. coli* O157:H7 strains produce different levels of phage and Stx2 upon incubation with a putatively susceptible *E. coli* strain, C600. Of note, phage from two *E. coli* O157:H7 clade 8 strains were the most infectious of those screened, while a *stx2c*-encoding phage possessed limited infectivity. Furthermore, our data suggested when coinoculating *E. coli* O157:H7 with susceptible *E. coli* in the absence of inducing antibiotics, the majority of Stx2 accumulation could occur due to lytic infection of the susceptible hosts rather than from the lysogen itself. Therefore, this study suggests that the intestinal microflora may dictate the severity and outcome of an O157:H7 infection, and toxin production observed in pure culture might not accurately reflect the levels produced *in vivo*.