

Identifying Conserved Signal Sequences in the Type III Secretion System of *Vibrio parahaemolyticus*

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The type III secretion system (T3SS) is an important virulence factor in many species of Gram-negative bacteria. T3SSs are used to translocate effector proteins into host cells where they induce changes to cellular physiology and modulate host-pathogen interactions. Effector proteins contain a signal sequence identifying them for T3SS secretion and/or translocation. Empirical studies indicate that the export signal is located within the first 30 amino acids from the N-terminus, but no consensus sequence or common structural motifs have yet been successfully identified. Some degree of signal conservation is expected to be present for T3SS recognition, as heterologous expression and successful T3SS secretion of effector proteins has been demonstrated, even in proteins originating from different genera. By coupling putative effector protein leader sequences from *Vibrio parahaemolyticus* with a phospholipase (YpIA), we identified 32 *V. parahaemolyticus* proteins that are secreted by the T3SS of the heterologous host *Yersinia enterocolitica*. We hypothesize that there is more than one conserved signal sequence for effector proteins and that different T3SS chaperones are involved in trafficking effectors based on their affinity for different export signals. We will test this hypothesis by first generating a mutant library of T3SS secretion signals using error-prone PCR. This data will allow identification of specific export sequences and subsequently allow identification of chaperone proteins involved in the export process. The data will be extended to identify effector proteins for other important gram-negative pathogens harboring a T3SS.