Computational modeling and experimental validation of the Legionella and Coxiella virulence-related type-IVB secretion signal

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AUTHOR SUMMARY

Many pathogenic bacteria use secretion systems that translocate a set of proteins, termed effectors, into their host cell. These effectors subvert various host cell processes for the benefit of the bacteria. Legionella pneumophila and Coxiella burnetii are two human intracellular pathogens that multiply in alveolar macrophages and use a type-IVB secretion system (1–3). These effectors have been shown to harbor a C-terminal secretion signal required for their translocation (4). This study aimed to characterize the secretion signal used by effectors to recognize the secretion system. We synthesized a functional artificial signal with no sequence similarity to any known effectors and discovered, on a genomic scale, effectors in related bacterial species for which no effectors had yet been identified. Hence, our study reveals important insights into the molecular mechanisms of bacterial pathogenesis.

To characterize the secretion signal of the Legionella and Coxiella effectors, we implemented a statistical method known as the hidden semi-Markov model (HSMM), which can account for dependencies among sites, and thus capture spatial variation in amino acid composition along the secretion signal. We used this model (i) to detect the existence of a signal on a genomic scale, (ii) to provide a quantification of the signal’s strength, and (iii) to measure the importance of specific amino acids at certain positions. Our findings indicate that an accurate description of the secretion signal should include more than just the existence of specific amino acids along the C terminus. Rather, our model suggests that efficient translocation depends on both the composition and spatial distribution of amino acids with similar physicochemical properties located along the 35 C-terminal amino acids. We validated our model by computing and synthesizing an artificial optimal secretion signal (Fig. P1A) and demonstrated that this signal can translocate a protein into host cells in a type-IVB-dependent manner (Fig. P1B). We next used our model to predict previously unidentified effectors in four Legionella species and in C. burnetii on a genomic scale (Fig. P1C). These predictions were experimentally examined in two pathogenic Legionella species, as well as in C. burnetii, resulting in the identification of a total of 20 effectors. Interestingly, some effectors received very low signal scores according to our model and yet were efficiently translocated. Our results suggest that this reflects the involvement of the IcmS–IcmW chaperon complex in their translocation.

Our findings uncover various ways by which the type-IVB pathogenesis system recognizes its substrates. Many other bacterial pathogens use secretion systems to translocate effector proteins into host cells; thus understanding the signal recognized by such systems is critical to revealing the function of these systems at the molecular level. Our approach also introduces a general experimental-computational methodology that can be used to identify a wide spectrum of protein features that lack sequence conservation but share similar amino acid characterics.


The authors declare no conflict of interest.

This article is a PNAS Direct Submission.


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Cite this Author Summary as: PNAS 10.1073/pnas.1215278110.