Structural and functional studies on Gram+ T4SS adhesion components

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In the recent years, infections caused by multi-drug resistant bacteria have become an increasing problem in healthcare systems all over the world. Spread of antibiotic resistance within a population is promoted by a highly effective system for the transfer of DNA, mediated by proteins of the Type IV Secretion System (T4SS) [1]. T4SSs are therefore a major contributor to the spread of antibiotic resistance in many clinically relevant pathogens [2]. An important step in this process is the formation of mating pairs between the donor and recipient cells. In Gram-positive (G+) T4SSs, adhesion of the cells is of particular interest, since there are no pili and thus adhesion proteins have to form mating cell aggregates and possibly also a pore in the cell wall of the receiving cell.

We study the pCF10 T4SS from *Enterococcus faecalis*, where at least three proteins have been identified to be directly involved in the adhesion process, named PrgA, PrgB and PrgC. It has been shown that the interaction of PrgA with PrgB is crucial for cell adhesion and that PrgB is a major contributor to *E. faecalis* virulence, while the function of PrgC is not yet elucidated.

We were able to purify, crystallize and solve the structure of different truncations of PrgA & PrgB at high resolution. The analysis of the structural data obtained for PrgA hint towards an unexpected function of that protein in the process of cell adhesion. Moreover, we report the structure of the adhesin domain of PrgB, which binds and strikingly also compacts DNA *in vitro*. *In vivo* PrgB deleted of its adhesin domain does not support cellular aggregation, biofilm development and conjugative DNA transfer, making it a potential drug target.

References

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