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Structures of major pilins in *Clostridium perfringens* demonstrate dynamic conformational change

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1 Introduction

Gram-positive bacteria possess a thick cell wall, a mesh polymer of peptidoglycans, that surrounds their cytoplasmic membranes and provides physical protection. The pathogenesis and infectivity of Gram-positive bacteria are mediated by many surface proteins that are covalently attached to the cell wall. Pili are flexible rod proteins associated with the bacterial cell surface, and play important roles in the initial adhesion to host tissues and colonization. The pilus shaft is formed by the covalent polymerization of major pilins. Here, we present X-ray structure of major pilin from *Clostridium perfringens* SM101 (CppA-SM101) and a structural comparison with major pilin from *Clostridium perfringens* strain 13 (CppA-St13) [1].

2 Experiment

Crystallization conditions of CppA-SM101 are as follows; protein concentration of 52 mg/ml, 4% v/v Tacsimate buffer pH 4.0 (73 mM malonic acid, 10 mM ammonium citrate tribasic, 5 mM succinic acid, 12 mM D,L-malic acid, 16 mM sodium acetate trihydrate, 20 mM sodium formate, and 6.4 mM ammonium tartrate dibasic), 12% w/v PEG3350. X-ray diffraction data were collected on the PF-BL5A beamline in KEK (Tsukuba, Japan), and were processed using the program XDS and the CCP4 program suite. The structure was solved by molecular replacement with the program MOLREP using the structure of CppA-St13 (PDB code: 5XCC). The structure was refined to R_{cryst} = 0.221 (R_{free} = 0.276) using 2.72 Å resolution data.

3 Results and Discussion

The structure of CppA-SM101 with 453 amino acid residues consists of three domains, D1, D2 and D3, and adopts a novel bent structure (Fig. 1A). In crystal, four molecules are associated to form a left-hand twist like an anti-parallel double helix, which likely promotes the bacterial cell-cell interactions with three possibilities: intra-chain, inter-chain, and inter-cell twists (Fig. 1B). While CppA-St13 adopts an elongated structure to form non-covalently linked polymeric chains in a crystal, yielding a practical model of the pilus fiber structure (Fig. 1A). Since amino acid sequence identity between them is 95%, CppA-SM101 was thought to be in an equilibrium state between an elongated and a bent structure through the dynamic conformational change (Fig. 1A), which may be involved in pili-mediated colonization.

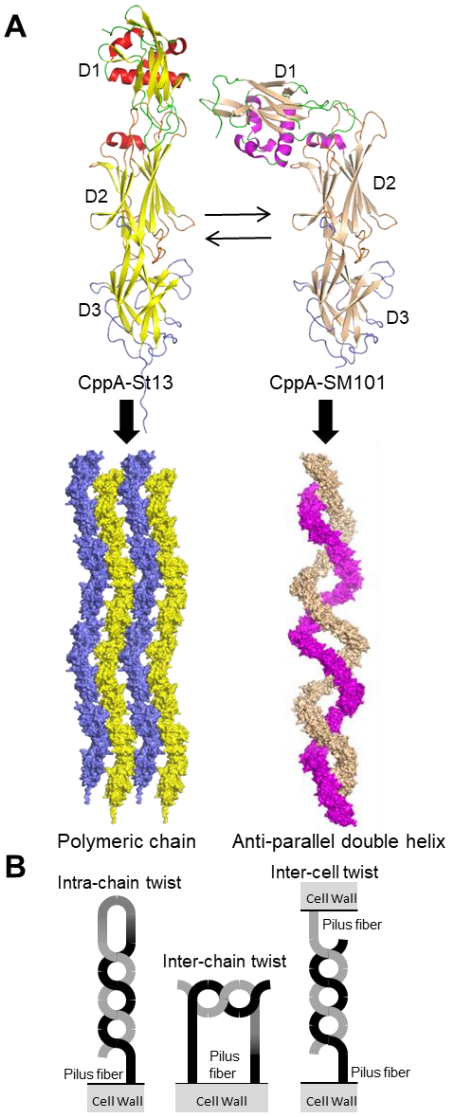

Fig. 1: Structures of CppA-St13 and CppA-SM101.

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References

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