The Mechanisms of Self-assembly and Polymorphic Switching of the Bacterial Flagellar Filament

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The bacterial flagellum is a helical filament, rotated by the motor at its base, and works as a screw that propels the cell. Yet, it is not a simply rigid propeller. The filament is normally in a left-handed, supercoiled form and several of them form a bundle when bacteria swim. Upon quick reversal of the motor rotation, the filaments switch into right-handed supercoils, making the bundle fall apart, enabling the cell to tumble for its tactic behavior. The filament is a tubular structure formed by self-assembly of single proteinflagellin in a helical manner. The supercoiling is thought to involve two distinct subunit conformations or packing, and its mechanism is interesting in terms of conformational distinctness and adaptability of flagellin. X-ray fiber diffraction and electron cryomicroscopy have been used to analyze the structures of various straight filaments. We developed a new method to orient liquid crystalline sols of filamentous macromolecular assemblies, by which the flagellar filaments have been aligned to 0.6 degree disorientation. X-ray diffraction from these specimens allowed us to measure the layer-line spacings, helical symmetries, and layer-line amplitudes accurately. With phases from the EM analysis, we obtained an electron density map at 9 Å resolution, which showed the packing of alpha-helices aligned along the protofilament in the core domain. About 65 N-terminal and 45 C-terminal residues of flagellin are disordered in the monomeric form, and proper interactions between the termini are essential for correct folding of these regions in the very inner core of the filament. Thus, these disordered portions are responsible for preventing spontaneous filament formation by monomers alone in the self-assembly mechanism. The structures of the L- and R-type straight filaments, which are thought to represent the two states of flagellin subunits that coexist in supercoiled filaments, showed only a small difference in the subunit packing and no appreciable differences in the overall subunit shapes. The intersubunit distance along the 11-stranded protofilaments is 52.7 Å and 51.9 Å for the L- and R-type, respectively; the L-type is longer than the R-type by 0.8 Å, quantitatively explaining the observed forms of supercoils based on a two-state subunit model. The conformational switching between the two states appears to be mutual sliding of the alpha-helical bundles in the core domain. The difference of 0.8 Å is produced by two distinct sliding distances, 1.8 Å and 2.6 Å, at two different intersubunit interfaces, respectively. These sliding movements are triggered by the shear force at the protofilament interface, which is produced by conversion of the twisting force by quick reversal of the motor rotation by the structure and packing of the flagellin subunits.