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Commentary and Perspective

Frontiers of microbial movement research

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Locomotion is one of the most fascinating aspects of living organisms, which is why various types of motor-protein complexes continue to fascinate many researchers around the world. To date, 18 different types of motility systems have been identified on Earth. Motor-protein complexes are highly dynamic and robust structures that convert electrochemical or chemical energy to mechanical action for movement. Interestingly, the motor-protein complexes autonomously adjust their mechanical functions in response to environmental changes. Many motility systems of microorganisms such as bacteria, archaea, and amoebae are under the control of sensory signal transduction networks, allowing microorganisms to migrate towards environments favorable for survival and away from unfavorable environments [1-3].

The 61st Annual Meeting of the Biophysical Society of Japan, to be held in November 2023, features a symposium with five invited speakers to discuss recent advances in the structure, assembly, and function of various microbial motility machines (Figure 1). This symposium also discusses in depth the molecular mechanisms behind diverse microbial motility processes and explores the design principles common to these seemingly diverse motility systems.

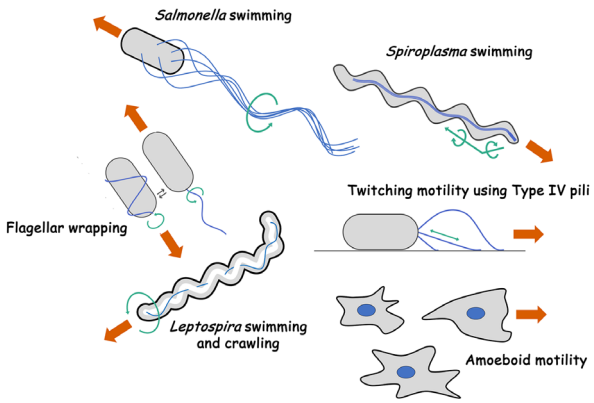

Figure 1 Various types of microbial motility systems discussed in this symposium

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Tohru Minamino at Osaka University reports on the proton-protein antiporter mechanism used in flagellar type III protein export in *Salmonella*. *Salmonella enterica* can swim in liquids and move on solid surfaces by rotating flagella. The *Salmonella* flagellum is a supramolecular complex consisting of a basal body, which acts as a bi-directional rotary motor, a hook, which functions as a universal joint, and a filament, which works as a helical propellor. Flagellar building blocks are transported via the flagellar type III secretion system (fT3SS) to the distal end of the flagellar structure. The fT3SS consists of a transmembrane export gate complex powered by proton motive force across the cytoplasmic membrane and a cytoplasmic ATPase ring complex [4]. ATP hydrolysis by the ATPase ring complex is postulated to transform an inactive export gate complex into a highly efficient protein transporter that couples inward-directed proton flow with outward-directed protein translocation across the cytoplasmic membrane [5-8]. In this symposium, he discusses how the fT3SS opens both proton and polypeptide channels to drive proton-coupled flagellar protein export.

Daisuke Nakane at the University of Electro-Communications reports on behavioral exhibition of bacteria. In recent years, there has been an increasing trend of behavioral exhibition in zoos. Animals are placed in environments that mimic their natural habitats, allowing visitors to observe their natural behaviors. This raises a simple question whether a similar approach can be taken with bacteria. To investigate this question, his research group has applied mechanical forces that are ubiquitous in the natural environment, such as water flow and spatial constraints, in a laboratory condition and has observed bacterial motility exposed to realistic environments under optical microscopy. In this symposium, he focuses on flagellar wrapping in a microfluidic device [9,10] and type IV pili-dependent rheotaxis [11,12] and discusses how these supramolecular machineries are controlled by the surrounding environments.

Shuichi Nakamura at Tohoku University reports on the biophysical aspect of a group of spiral-shaped bacteria called spirochetes. The bacterial flagella of most species extend to the cell exterior, whereas spirochetes possess their flagella within the periplasmic space. The periplasmic flagella of spirochetes transform or rotate their spiral or wavy cell bodies. The spirochete flagella are located at both poles of the cell body, which is approximately 20 μm long. Physical insights suggest the significance of synchronous operation of the flagellar motors up to 20 μm apart, but no convincing evidence has been presented. As is known for many motile pathogens, spirochete motility seems likely to be involved in pathogenicity. In this symposium, he discusses the mechanism by which spirochete migrates using bipolar periplasmic flagella and its role as virulence factors [13-15].

Hana Kiyama at Osaka Metropolitan University reports on the mechanism of minimal cell motility using bacterial actins reconstructed in a synthetic minimal bacterium. *Spiroplasma*, wall-less helical bacteria, can swim in liquids by switching their helicity between left-handed and right-handed using an intra-cellular ribbon structure containing five distinct bacterial actins named MreB1, MreB2, MreB3, MreB4, and MreB5. Her research group has successfully reconstructed the helical morphology and the swimming motility of *Spiroplasma* in the synthetic minimal bacterium JCVI-syn3B by expressing MreB4 and MreB5 together, but the underlying mechanism by which *Spiroplasma* cells swim is still unknown [16]. To clarify this mechanism, her research group induce mutations into these MreB proteins and analyzes the behavior of the synthetic minimal bacteria expressing the mutated MreB proteins. In this symposium, she describes the importance of the membrane binding region of MreB5 at the C-terminus on the swimming and the ATPase activity of MreB4 on the helicity switching from right to left.

Yusuke V. Morimoto at Kyushu Institute of Technology reports on visualization of signal transduction in unicellular and multicellular stages of *Dictyostelium*. The cellular slime mold *Dictyostelium discoideum* is a model organism for cell motility, including chemotaxis, and collective cell migration. *Dictyostelium* cells periodically produce and release cAMP as a signal upon starvation and aggregate to form multicellular bodies in a chemotactic manner. His research group has visualized the signals in each developmental stage of *Dictyostelium* cells. Live cell imaging of cytosolic cAMP reveals that oscillations of the cAMP signal are clearly observed during the cell aggregating stage, but the periodic signals gradually disappear during the multicellular body formation stage [17]. On the other hand, Ca^{2+} and c-di-GMP signals are visualized as signals that function in the multicellular stages [18,19]. In this symposium, he discusses how these chemical signals regulate multicellular body formation.

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