Investigation of a potential force-generation machinery driven by a cytoskeletal Walker-type ATPase in prokaryotic cells

ANDREAS ERBE, SING-YI HOU, CHEN-YUN CHEN, YI-LIH LIN, JIE-PAN SHEN, LI-JING LIN, CHIA-FU CHOU, YU-LING SHIH, Academia Sinica — Cytoskeletal proteins are often involved in generating mechanical force to drive various cellular processes. A subgroup of the Walker-type ATPases acts as cytoskeletal proteins that show highly dynamic behavior in bacterial cells. One of the most prominent examples is MinD that works with other cellular components to prevent cell division at unwanted polar sites through cycles of pole-to-pole oscillation in *E. coli* cells. We use fluorescence microscopy techniques to study the process of MinD assembly and disassembly on a lipid bilayer membrane surface and any possible change of membrane properties caused by MinD association with the membrane. To form a supported bilayer membrane, vesicles of the polar or total extract of *E. coli* membrane or synthetic lipids of defined composition are adsorbed to a treated glass coverslip. Ca$^{2+}$ is added to enable vesicle fusion to form a continuous bilayer on a glass surface. Formation of a bilayer is examined using fluorescence recovery after photobleaching. The results on the protein assembly on membranes present an important step in understanding the intermediate stages that occur during the dynamic movement of MinD in cells.

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