3P199 Ion selectivity of the *Leptospira* flagellar motor

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Leptospira have intracellular flagella that rotate within the periplasmic space. Each flagellar filament is linked to a flagellar motor. Although the bacterial flagellar motor is powered by the electrochemical potential gradient of specific ions across the cytoplasmic membrane, coupling ions driving the *Leptospira* motor has not been investigated. We tested effects of a sodium channel inhibitor, EIPA, and a protonophore, CCCP, on the motility of saprophytic *Leptospira biflexa*, showing that the motility is completely inhibited by EIPA but not by CCCP. We also found that the motility is significantly enhanced with concentrations of NaCl added to media. These results suggest that the flagellar motor of *Leptospira* can utilize Na⁺ as a coupling ion for torque generation.

3P200 Functional and structural analysis of the flagellar protein FliL from *Vibrio alginolyticus*

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The bacterial flagellum consists of many different proteins. Although the functions of most of these proteins have been characterized, there are a few which remains to be studied in detail. FliL is one such enigmatic integral membrane protein. To clarify the function of FliL, we deleted fliL gene from chromosome of *Vibrio alginolyticus*. Pilot experiments showed that motility phenotype of the mutant was not different from that of the wild type. Recombinant full length FliL and its periplasmic region were over-expressed in *Escherichia coli*, purified and attempts are being undertaken to crystallize the purified proteins in order to solve its structure. Studies are also being carried out to understand its localization and possible interaction with other flagellar proteins.

3P201 Actin filament dynamics and organizations in liposome: A simulation study

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Dynamics and organizations of actin filaments play essential roles in cell morphology and motility. The dynamics and organizations were known to be affected by associated proteins and surrounding boundaries (cell membranes). However, complication in cellular environments has hampered quantitative and systematic investigations on what are essential ingredients for phenomena of interests. Reconstructed systems, actin filaments and associated proteins in liposomes, has enabled to investigate the dynamics and organizations with well-defined components, enabling direct comparison with simulations. Here, with a computer simulation, we will present a systematic investigation on dynamics and organizations of actin filaments in liposomes.

3P202 海洋性ビブリオ菌のべん毛形成を制御する DnaJ ファミリータンパク質 SflA の相互作用解析

Analysis of interaction of the DnaJ family protein SflA, that is involved in regulation of flagellation in *Vibrio alginolyticus*

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Marine bacterium *Vibrio alginolyticus* usually has a single polar flagellum, and its number and location are regulated by FlhF and FlhG. Mutation in *sflA* restores motility of the severely reduced $\Delta flhFG$ mutant, and the single transmembrane protein SflA was found to be located at the cell pole. We speculated that SflA interacts with FlhF, FlhG and other proteins, and suppress the flagellation. Function of the large N-terminal periplasmic region of SflA is not well understood, and its cytoplasmic region may interact with FlhF and FlhG. In order to test our hypothesis, we investigate binding protein(s) of SflA by the pull down assay using N- or C-terminal fragment of SflA. Our recent results about crystallization will be also presented.

3P203 非対称な脂質 2 重膜の高効率な作成にむけた新規マイクロデバイスの開発

Novel micro-device to form asymmetric lipid-bilayer membrane in a high throughput manner

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Most biological membranes possess an asymmetric transbilayer distribution of phospholipids. In particular, a plasma membrane of eukaryotes maintain a high asymmetry, controlling various cellular functions. To understand them, extensive studies has been performed to develop artificial lipid-bilayer systems; however, it remains difficult to form asymmetric lipid-bilayers in a high-throughput manner. In this study, we developed a novel micro-device to form 10,000 asymmetric lipid-bilayers at a time with high efficiency of 97 %. This micro-device extends the versatility of artificial lipid-bilayer system, which had been in general limited to symmetric-bilayer formation, and will contribute to the understanding of the role of lipid compositional asymmetry on cell physiology.

**3P204 脂質キュービック相の単結晶化
Single Crystallization of an Inverse Bicontinuous Cubic Phase of a Lipid**

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We report a simple method to produce a single crystal region of an inverse bicontinuous cubic (Q_{II}) phase of a lipid, 1-monolein. By starting with the lipid of the sponge (L₃) phase in the presence of 1,4-butanediol, we can obtain a single crystal region of the double-diamond Q_{II} phase in one week by controlled dilution of 1,4-butanediol. The length of the single crystal region in a 0.5 mm diameter capillary was on the order of mm. X-ray diffraction images changed rotation angle dependently. We could assign Miller indices to all of the distinguishable diffraction spots from the region. This method would bring benefits to the basic and application researches of the Q phases.