2P193 ケージドセリン光分解とべん毛モーター回転計測を用いた大腸菌走化性応答の高時間分解能計測

Response of flagellar motor rotation to photoreleased serine from caged-compound in an *E. coli* cell

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E. coli cell controls rotational direction of flagellar motor by chemotaxis system. To understand the signaling process in high-temporal resolution, a cellular response time and duration of response to photoreleased serine from caged-compound were measured via the rotational direction of a motor. From the response time and the duration of response, the same values of threshold were estimated in the increase and decrease of serine-concentration, indicating a cell senses absolute value of serine-concentration. Moreover, we found out the response time depends on the distance between receptor and motor. This dependency in response time informed us that diffusion-based propagation of signaling molecules and the 240 ms of enzymatic reaction involved in the signaling process.

2P194 MS リングに変異の入ったサルモネラ菌べん毛モーターとそのシュードリバータントの構造安定性と回転特性

Structural stability and rotational characteristics of the flagellar motor of Salmonella MS-ring mutant and its pseudo-revertants

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The bacterial flagellar motor is a rotary nanomachine fueled by proton motive force. It has been known that one of *Salmonella* FliF mutants has fragile flagella as compared with those of a wild type strain. In this study, we quantitatively measured the mechanical strength and torques of the wild-type and FliF mutant flagella. As a result, the strength of the FliF mutant decreased to a half of the wild type. Several pseudo-revertants of the FliF mutant suppressed fragility of the flagella. In contrast, rotation assays revealed that motor torques of the FliF mutant and pseudo-revertants were almost the same with those of the wild-type at high loads. These suggest that the mutation in FliF decreases structural stability of the motor but does not affect torque generation.

2P195 電子顕微鏡によるヒト毛乳頭細胞の不動毛の構造解析
Structural analysis of primary cilia in human follicle dermal papilla cells by electron microscopy

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Most cells of multicellular organisms have “primary cilia”, which are single, non-motile, and sensory cilia. They have been reported to detect and transform mechanical stimulation in cell. Dermal papilla cells (DP cells) with primary cilia (Matsushima, 2009) in the skin can induce hair growth. Their ultrastructure is, however, unclear. Here we observed the structure of primary cilia of DP cells by confocal laser microscopy and scanning electron microscopy. As a result, we, for the first time, observed the 80 % primary cilia of the cells around the outside of the nucleus, which is ~2.9 μm in length and 200 nm in diameter. We believe that these results help to reveal the function of primary cilia of DP cells.

2P196 簡単に低コストなコラティブ顕微鏡法

Simple and cost-effective method for correlative microscopy

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Recently it has been growing a demand to observe cytoskeleton in a locomoting cell at molecular level. The methods combining a live cell imaging to an electron-microscopic observation have been reported as correlative microscopy requiring hi-end equipments. Here we show a simple and cost-effective method for the correlative microscopy. BG2-c6 cells, *Drosophila* neural cell line, were cultured on formvar film attached on the finder grid. After fixation and extraction of cells with the glutaraldehyde and detergent mixture at the end of live imaging, the finder grid was picked up to negative-stain cells. Under a transmission electron microscope, cells of interest were located by the specific pattern of the grid to observe the fine structure at high magnification.

2P197 顕微鏡ステージ上での微量エレクトロポレーション法

Electroporation of adherent cells with low sample volumes on a microscope stage

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Electroporation is a powerful technique to label specific molecules in living cells for investigating intracellular molecular dynamics. However, the loading of samples into “adherent” cells with normal electroporators is difficult. Here, we developed a new electroporator with four special characteristics: (1) Electric pulses are applied to the adherent cells directly, without removing them from the substratum. (2) Samples can be loaded into the adherent cells while observing them on the stage of an inverted microscope. (3) Only 2 μl of sample solution is sufficient. Samples could be loaded into keratocytes, neutrophil-like HL-60 cells and *Dictyostelium* cells. The new device should be useful for a wide range of adherent cells.

2P198 神経一瞬島 α 細胞相互作用におけるサブスタンス P の寄与
A neuropeptide substance P is involved in nerve-pancreatic islet α cell interaction

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Autonomic neurons innervate pancreatic islets of Langerhans and maintain blood glucose homeostasis by regulating hormonal levels. Using in vitro coculture of αTC6 cells, a murine islet α cell line, with superior cervical ganglia (SCG), we observed the oscillation of intracellular Ca²⁺ concentration in αTC6 cells after SCG activation specifically with scorpion venom. In addition, we found that the neurokinin (NK)-1 receptor, a substance P receptor, was expressed in αTC6 cells, and that pretreatment with CP99,994, an NK1 receptor antagonist, suppressed the responding rate of αTC6 cells. These results suggested that substance P released from stimulated neurites functioned as a mediator to activate α cells.