The critical role of electrostatic interaction in actin-myosin dissociation inferred from ATP-induced allosteric responses of myosin

Takato Sato, Jun Ohnuki, Mitsunori Takano (Grad. Sci. of Adv. Sci. & Eng., Waseda Univ.)

In muscle contraction, myosin moves along actin filament repeating the strong and weak binding alternately. Although this strong/weak binding transition is essential for the cyclic energy-conversion of myosin, the atomic-level understanding of this process has not yet been established. The ATP-binding to myosin triggers the dissociation from the actin, and as a possible mechanism, opening of the actin-binding cleft is speculated. However, other structural elements of myosin including loop 2 and surrounding solvents have received little attention. In this study, to clarify the allosteric responses of the structural and the solvation states of myosin to strong/weak binding transition with atomic details, we conducted explicit-solvent molecular dynamics simulation. The crystal structure of scallop myosin was used, and the chemical state was changed from the original ADP-Pi analogue bound to the ATP bound or nucleotide-free state. Then, myosin was immersed in the box containing 33,000 waters and 50 mM KCl. From multiple long MD runs (total over 6 μs), the anticipated structural responses in the actin-binding cleft and loop 2 were observed. Furthermore, density increase of water and exclusion of anion at around the actin-binding interface upon ATP-binding were observed. It is likely that these responses are explained by the change of electrostatic potential around myosin upon ATP-binding. These "allostery of structure and solvent" of myosin is expected to be deeply involved in the dissociation of myosin from the actin filament.

3M1334  生体外でのアクチノミオン運動型リポソーム

Actomyosin-driven liposomes in vitro

Satoshi Iwabuchi, Kaniyuki Hatori (Grad. Sch. Sci. Eng., Yamagata Univ.)

We intended to add a function of active motion to liposomes by actomyosin motor proteins. Using a hydration method, positively-charged liposomes were prepared from a equimolar mixture of dioleoyl-phoshatidylcholine (DOPC) such as a neutral lipid with dioleoyl-trimethyl ammonium propane (DOTAP), which is a cationic lipid. When the mixture of the liposomes and rhodamine-phallolidin-bound actin filaments was observed under a fluorescent microscope, it was found that actin filaments were bound to the surface of liposomes. It is likely that the binding of actin filaments to liposome surface was caused by electrostatic interaction. Under an in vitro motility assay, we observed that the actin-bound liposomes moved at about 1.2 μm/s of velocity on heavy meromyosin (HMM) molecules fixed on glass surface in the present of ATP. This motion was different from Brownian motion, and was slower than sliding movement of actin filaments without liposome on HMM (about 5 μm/s). We achieved introduction of actomyosin system into liposomes for active transport.

3M1346  Characterization of photochromic ATP analogue as a substrate for myosin

Takeshi Itaba, Shinsaku Maruta (Div. of Bioinfo., Grad. Sch. of Eng., Soka Univ.)

Azobenzene is a photochromic molecule that undergoes rapid and reversible isomerization between the cis- and trans-forms in response to ultraviolet and visible light irradiation, respectively. Previously, we have cross-linked reactive cysteine SH1 and SH2 of skeletal muscle myosin subfragment-1(S1) with the sulfhydroxyl-reactive bifunctional azobenzene derivative, ABDM and succeeded to induce lever arm swinging reversibly by photo-irradiation. The results suggested that it may be possible to control motor proteins by light irradiation.