Atlas of Science
another view on science
https://atlasofscience.org

Dynamic electron microscopy: recording of ATP-induced myosin head movement in living muscle myosin filament

Muscle contraction results from relative sliding between actin and myosin filaments, caused by cyclic movement of myosin heads coupled with ATP hydrolysis. It is generally believed that individual myosin heads M), extending from myosin filaments, first bind with actin filaments (A) in the form of M-ADP-Pi to perform power stroke producing unitary filament sliding, associated with reaction, M-ADP-Pi + A → A-M-ADP-Pi →A-M + ADP + Pi. After completion of Power Stroke, M detaches from A by binding with ATP, and perform recovery stroke to return to their initial position as, A-M + ATP → A + M-ATP →A-M-ADP-Pi. The ATP-induced myosin head strokes, however, still remain to be a matter for debate and speculation.

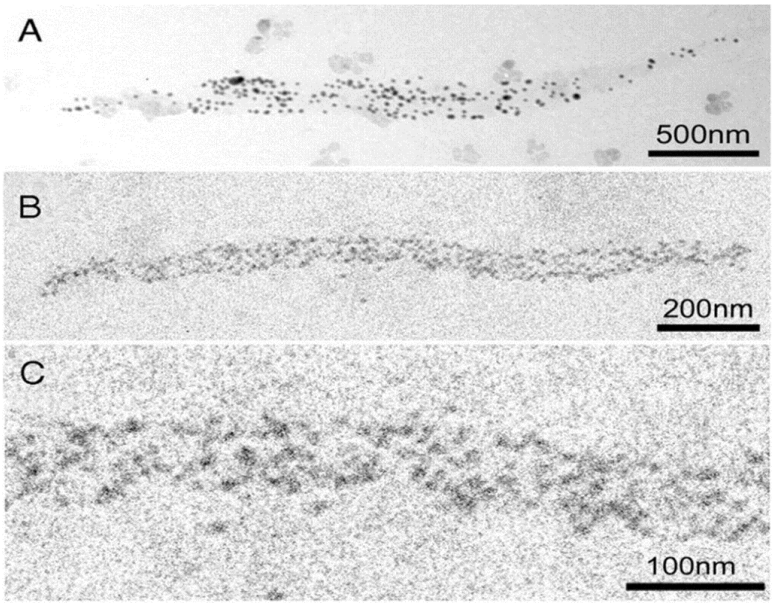

Fig. 1. Electron micrographs of spindle-shaped synthetic myosin filaments.

By using the gas environmental chamber (EC, or hydration chamber), which enables us to visualize and record dynamic structural changes of living biological biomolecules under an electron microscope, we have succeeded in recording ATP-induced movement of individual myosin heads in wet, living synthetic myosin filaments, prepared from rabbit skeletal muscle (magnification, 10,000X). Individual myosin heads were position-marked by attaching gold particles (diameter, 20nm) via a monoclonal antibody to the distal region of myosin head. The filament images were recorded with an imaging plate system (Fuji Photofilm, Tokyo, Japan). Figure 1 shows electron micrographs of spindle-shaped synthetic myosin filaments, in which individual myosin heads are position-marked with gold particles. Each gold particle image consisted of many dark particles. We determined the center of mass position of each gold particle, and it was taken as the position of each myosin head. The myosin head position remained unchanged without ATP application. The spindle shape of myosin filaments arises from symmetrical arrangement of myosin molecules with respect to the bare region located at the middle of myosin filament, across which the myosin head polarity is reversed, so that myosin head power and recovery stroke takes place towards and away from the bare region, respectively.

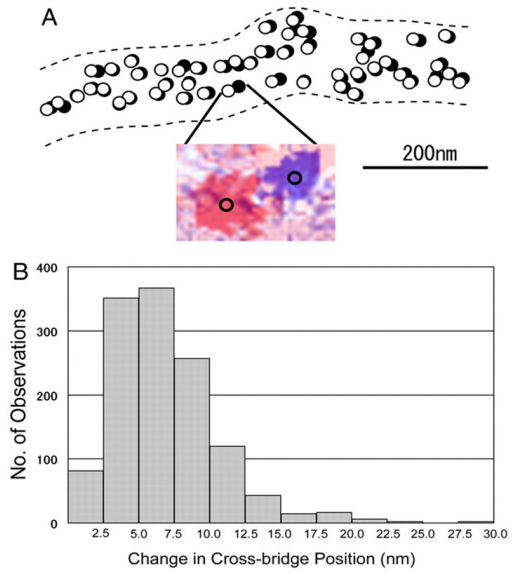

Fig. 2.

We applied ATP to myosin heads In the absence of actin filaments, by passing current to an ATP-containing microelectrode (Sugi et al. Proc Natl Acad Sci USA, 105:17396-17401, 2008): In Figure 2A, positions of individual myosin heads before and after ATP application are indicated by open and filled circles (diameter, 20nm), respectively. On ATP application, individual myosin heads were found to move in one direction along myosin filament long axis. As shown in Figure 2B, the average amplitude of ATP-induced myosin head movement was ~6nm. After complete exhaustion of ATP, individual myosin heads returned to their initial position. At both sides of the bare region at the middle of myosin filament, myosin heads were found to move away from, but not towards the bare region, indicating that, in the absence of actin filament, myosin heads exhibit recovery stroke with direction opposite to that of power stroke. This finding indicates that myosin heads can decide their movement direction without being guided by actin filaments. Finally, we emphasize that the dynamic electron microscopy, which is created by us, is a powerful tool to open new horizons in the research field of biology and medicine.

Haruo Sugi
Department of Physiology, Teikyo University School of Medicine, Tokyo, Japan

Publication

Direct demonstration of the cross-bridge recovery stroke in muscle thick filaments in aqueous solution by using the hydration chamber.
Sugi H, Minoda H, Inayoshi Y, Yumoto F, Miyakawa T, Miyauchi Y, Tanokura M, Akimoto T, Kobayashi T, Chaen S, Sugiura S
Proc Natl Acad Sci U S A. 2008 Nov 11