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Electron microscopic visualization and recording of myosin head recovery and power strokes in hydrated myosin filaments using the gas environmental chamber

Although more than 50 years have passed since the monumental discovery of sliding filament mechanism in muscle contraction, the molecular mechanism of myosin head movement, coupled with ATP hydrolysis, is still a matter for debate and speculation. Most straightforward way to study myosin head movement, producing myofilament sliding, may be to directly record ATP-induced myosin head movement in hydrated, living myosin filaments using the gas Environmental Chamber (EC) attached to an electron microscope. While the EC has long been used by materials scientists for the *in situ* observation of chemical reaction of inorganic compounds, we are the only group successfully using the EC to record myosin head movement in living myosin filaments. We position-mark individual myosin heads by attaching gold particles (diameter, 20 nm) via three different monoclonal antibodies, attaching to: At the distal region of myosin Head Catalytic Domain (CAD); at the myosin head Converter Domain (COD) and at the myosin head Lever arm Domain (LD). First, we recoded ATP-induced myosin head movement in the absence of actin filaments and found that myosin heads moved away from the central bare region of myosin filaments. This finding constitutes the first direct electron microscopic recording of myosin head recovery stroke under a condition in which myosin heads almost freely with average amplitude of ~7 nm. After many efforts, we succeeded in recording ATP-induced myosin head power stroke in actin-myosin filament mixture in 2015. Since only a limited proportion of myosin heads can be activated by a limited amount of ATP applied, myosin heads only move by stretching adjacent sarcomere structures, i.e., nominally isometric condition. Myosin head CAD did not move parallel to the filament axis in the standard ionic strength, while it moved parallel to the filament axis at low ionic strength, in accordance with our physiological experiments on single muscle fibers. These results indicate that myosin head movement does not necessarily obey predictions of the swinging lever arm hypothesis appearing in every textbook.

Biography

Haruo Sugi has received Post-graduate degree in the University of Tokyo and PhD, and was appointed to be an Instructor in Physiology in the University of Tokyo Medical School. From 1965 to 1967, he had worked at Columbia University as Research Associate and at the National Institutes of Health as a Visiting Scientist. He was the Professor and Chairman in Teikyo University Medical School from 1973 to 2004, when he became Emeritus Professor. He was also the Chairman of Muscle Commission in the International Union of Physiological Sciences from 1998 to 2008 (IUPS).

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