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Haruo Sugi

Teikyo University School of Medicine, Japan

Electron microscopic recording of ATP-induced myosin head power and recovery strokes in hydrated muscle myosin filaments using a film-sealed hydration chamber

uscle is a machine converting chemical energy derived from ATP hydrolysis into mechanical work. Although it has been well Lestablished that muscle contraction is produced by relative sliding between actin and myosin filaments, which in turn is caused by repeated myosin head power and recovery strokes, despite extensive studies over more than 20 years, the myosin head movement coupled with ATP hydrolysis still remains to be a matter for debate and speculation. As early as 1980s, we started to visualize ATPinduced myosin head movement in hydrated myosin filaments under a 200kV transmission electron microscope, using a film-sealed hydration chamber developed by Fukami in Nihon University. After a number of trials and errors, we have succeeded in recording ATP-induced myosin head movement, position marked with antibodies to myosin heads, in both the absence and the presence of actin filaments with the following results. Without ATP application, the time-averaged position of individual myosin heads do not change appreciably with time, suggesting that myosin heads undergo thermal fluctuation around a stable equilibrium position. In the absence of actin filaments, individual myosin heads move freely (average amplitude, ~7 nm) in response to iontophoretically applied ATP away from the bare region at the center of myosin filaments, indicating that the ATP-induced myosin head movement corresponds to myosin head recovery stroke. In the presence of actin filaments, individual myosin heads move by ~3.3nm (average amplitude, ~3.3 nm at the distal region and ~2.4 nm at the proximal region). The small amplitude of myosin head power stroke results from that only a small fraction of myosin heads are activated by ATP, so that individual myosin heads move for a limited distance by pulling adjacent elastic structures. At low ionic strength, the amplitude of ATP-induced myosin head power stroke increased to >5 nm in both distal and proximal regions, being consistent with our physiological finding that the force generated by individual myosin heads increases nearly two fold. We emphasize that the film-sealed hydration chamber coupled with iontophoretic ATP application is a powerful tool in solving many mysteries in biological systems.

Biography

Haruo Sugi has graduated from Postgraduate School of Life Sciences at the University of Tokyo with PhD degree in 1962 and appointed to be Instructor in Physiology at the University of Tokyo. From 1965 to 1967, he stayed in USA as a Research Associate at Columbia University and as a Visiting Scientist in NIH. In 1973, he became Professor and Chairman in the Department of Physiology, Teikyo University. From 1994 to 2006, he was the Chairman of Muscle Commission in the International Union of Physiological Sciences. In 2004, he has retired from his position in Teikyo University and he is currently an Emeritus Professor of Teikyo University.

sugi@kyf.biglobe.ne.jp