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Direct recording of myosin head power and recovery strokes in hydrated myosin filaments provides evidence against the swinging lever arm mechanism in muscle contraction

Muscle contraction results from relative sliding between actin and myosin filaments, which in turn is caused by cyclic attachment and detachment between myosin head extending from myosin filaments and active sites on actin filaments. A myosin head consists of catalytic (CAD), converter (COD), and lever arm (LD) domains, and connected to myosin filament backbone via subfragment-2. Based on crystallographic and electron microscopic studies on static structures of myosin heads and acto-myosin complex, it has been proposed that myosin head exerts power stroke by active rotation of CAD around CD, coupled with ATP hydrolysis. This mechanism is called "swinging lever arm mechanism", and now appears in every textbook as a dogma explaining molecular mechanism of muscle contraction. Using the gas environmental chamber, in which hydrated biomolecules can keep their function in the electron microscope, we succeeded in recording ATP-induced power and recovery strokes of myosin heads, which are position-marked with two different antibodies, attaching to junctional peptide between 50k and 20k segments of myosin heavy chain in CAD(antibody 1), and to reactive lysine residue in COD (antibody 2), respectively. Although antibody 1 covers two main myosin-binding sites on actin to inhibit formation of actin-myosin linkages, it has no effect on both Ca^{2+}-activated muscle fiber contraction and in vitro actin-myosin sliding. On the other hand, antibody 2 shows no effect on muscle fiber contraction, but completely inhibits in vitro actin-myosin sliding. These findings, together with our success in recording power stroke of myosin heads position-marled with antibodies 1 and 2, constitute evidence against the dogma (or textbook view) that (1) during muscle contraction, myosin heads do not pass through rigor configuration, and (2) muscle contraction does not results from active rotation of CAD around COD.

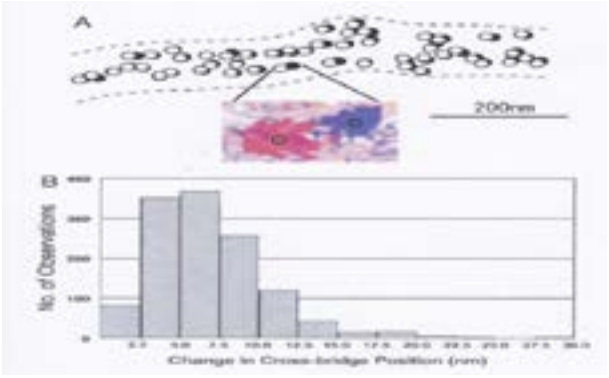

Figure 1: *ATP-induced myosin head recovery stroke in the absence of actin filament. Open and filled circles (diameter, 20nm) show the position of gold particles, attached to myosin heads with antibody 1) before and after ATP application, respectively. (Inset) Enlarged view showing the position of gold particle before (red) and after (blue) ATP application. (B) Histogram showing amplitude distribution of ATP-induced myosin head recovery stroke.*

Biography

Haruo Sugi graduated from postgraduate School in the University of Tokyo, Japan, with a PhD degree in 1962, and was appointed instructor in the Department of Physiology in the University of Tokyo. From 1965 to 1967, he worked at Columbia University as a research associate, and at the National Institutes of Health as a visiting scientist. He was a professor and Chairman in the Department of Physiology, Teikyo University Medical School, Japan, from 1973 to 2004, when he became an emeritus professor. Sugi was also chairman of the muscle committion in the International Union of Physiological Sciences (IUPS) from 1998 to 2008.

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