A Low Angle Diffraction Study of the Effects of Nucleotide Binding on the Structure of the Actomyosin Complex	X9B

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With an atomic model for the actin filament (Holmes et.al. 1990, Nature, 347: 44-49; Lorenz et. al. 1995, J. Mol. Biol. 246: 108-119) and the atomic coordinates for at least one form of the myosin motor protein (Rayment et.al. 1993, Science 262: 50-58) we are in a position to make use of high angle fibre diffraction data from myosin decorated actin filaments to refine a model structure of the complex. Nucleotide-free myosin subfragment-1 (S1) isolated from both chicken skeletal and smooth muscles was introduced into overstretched rabbit psoas muscle fibres. In both cases the protein binds tightly to actin and this results in the intensification of the actin based layer lines in the X-ray pattern. Refining an actomyosin atomic model against the nucleotide-free skeletal S1 data produced a best fit which was similar to the model suggested by Rayment et. al. (1993) based on electron microscope evidence. ADP binding had very little effect on this diffraction pattern. The nucleotide-free (unphosphorylated or thiophosphorylated) smooth muscle S1 decorated filaments produced a very different diffraction pattern and the binding of ADP was shown to induce a structural change in the head resulting in a pattern similar to that from skeletal S1 decorated fibres. The Kd for ADP binding was shown to be ca. 2 micromolar.

It is most interesting that the two rigor complexes are structurally different. If the rigor structure represents the end of the power stroke then the data imply that at least some part of the power stroke in these two motors is produced by different configurational changes of the actomyosin complex. Alternatively, we must question our interpretation of the rigor structures themselves. We are in the process of refining the model against the smooth muscle S1 data but it is clear that the ADP induced structural change in the myosin head is very large and certainly involves the distal part of the head. This may reflect the tail wagging motion described by Whittaker et. al. 1995, Nature 378: 748-751, from their electron microscopic evidence.

Data were collected on Fuji imaging plates at beamlines X9B at the NSLS, Brookhaven and BL4, ID2 at the ESRF, and scanned using a Fuji BAS2000 system.