Abstract Submitted
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Twirling of Actin by Myosins II and V JOHN F. BEAUSANG, Phyiscs Dept., HARRY W. SHROEDER, Biochemistry and Mol. Biophysics, JAMES A. GILMOUR, Dept of Bioengineering, YALE E. GOLDMAN[1], Pennsylvania Muscle Institute — A polarized total internal reflectance fluorescence microscope was used to measure the 3D orientation of single rhodamine molecules with 40 ms time resolution (Forkey et al., *Nature* 422:399, 2003). We modified this setup by adding excitation polarizations at $\pm 45°$ relative to the optical axis of the microscope, thus enabling us to uniquely monitor 1/4 of the probe's angular phase space (previously 1/8). Phase shifts in the optics were compensated with an adjustable waveplate. Using actin filaments sparsely labeled with tetramethylrhodamine at Cys-374, the increased range of discernable angles was used to determine the handedness of filament rotation about its axis in a gliding filament assay. During translocation by Myosin II or V, approximately half of the observed actin filaments exhibit a 'twirling' helical path of rotation around the filament axis. Myosin II and V consistently induce a left-handed twirling motion (opposite to the long-pitch helix of actin) with pitch of $1.0 \pm 0.2\,\mu$m for myosin II and $1.5 \pm 0.1\,\mu$mfor myosin V. Several factors may be the cause of this twirling including: the direction of the force vector between actin and myosin, the distribution of myosin binding sites on actin, and cooperation between myosins translocating an individual actin filament. Supported by NIH grant AR26846 and NSF grant DMR04-25780.

[1]University of Pennsylvania

John F. Beausang
Phyiscs Dept.

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