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Track Switching and Crossing by Microtubule Motors. JENNIFER ROSS, KAREN WALLACE, HENRY SHUMAN, ERIKA HOLZBAUR, YALE GOLDMAN, Pennsylvania Muscle Institute, University of Pennsylvania — Cytoskeletal filaments in cells form a network of crossing tracks for motor proteins carrying vesicular and protein cargoes. The ability to pass through, switch, or dissociate at such intersections is relevant to the motor's ability to effectively navigate in the cell and deliver goods to the appropriate location. We have formed an *in vitro* system of crossed microtubules to study the outcome of kinesin motors and dynein-dynactin complexes when they encounter an intersection. Microtubules were flowed into the sample chamber from two orthogonal directions and aligned with the flow direction when they attached to glass cover slips via biotin-streptavidin. The first flow direction defined the microtubules closest to the glass surface. Using total internal reflection fluorescence (TIRF) microscopy, we visualized single GFP-kinesin motors and dynein-dynactin-GFP complexes during processive motility at 1 mM ATP. Both dynein and kinesin can switch microtubules, pass by an intersection, or dissociate. Using optical trapping, we placed 1 μm polymer beads decorated with multiple motors to simulate a large cargo encountering an intersection at 1 mM ATP. Beads are more likely to pause at the intersection at high motor number and can pass and switch as the motor concentration is titrated down. The differences between kinesin and dynein could inform of the ability of these motors to navigate the cell, both separately and in coordination. Supported by NIH grant AR51174.

Jennifer Ross
Pennsylvania Muscle Institute, University of Pennsylvania

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