

USING LIGHT TO DISSECT AND DIRECT THE ORGANIZATION AND TRANSPORT WITHIN CELLS

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The organization and dynamics of the MT (MT) cytoskeleton underlies the morphology, polarization and division of most cells. The structural polarity of MT determines the directionality of motor proteins, which move selectively towards either the MT plus (most kinesins) or minus end (dynein) to control the transport and positioning of proteins and organelles. Our research is aimed at understanding how different cellular MT arrays, and in particular neuronal MT networks, are built and utilized to ensure proper cellular logistics. In this lecture, I will highlight recent developments from our group that use novel types of light-based microscopy to unravel the traffic rules and the road map within cells.

To explore how different motor proteins contribute to intracellular transport and to study the site-specific roles of different organelles, we have established both chemical and optical control of intracellular transport by using either ligand-induced or light-sensitive heterodimerization to recruit specific cytoskeletal motor proteins (kinesin, dynein or myosin) to selected cargoes. Motility of selected organelles is rapidly induced and can be controlled with spatiotemporal precision. Furthermore, we have also established multi-color control, enabling us to independently recruit different motors to different cargoes.

In neurons, these type of experiments have revealed that different plus-end directed kinesin have different preferences, moving either exclusively into axons or targeting both axons and dendrites. To unravel how the specialized organization of the neuronal cytoskeleton guides different motor proteins to either axons or dendrites, we have developed novel approaches for optical nanoscopy. One of these, called motor-PAINT, uses nanometric tracking of motor proteins to super-resolve cytoskeletal fibers and determine their polarity. This has revealed a key architectural principle of the neuronal microtubule cytoskeleton that explains how different motor proteins can selectively transport cargoes to either axons or dendrites.