Studying Microtubule Dynamics with Optical Tweezers

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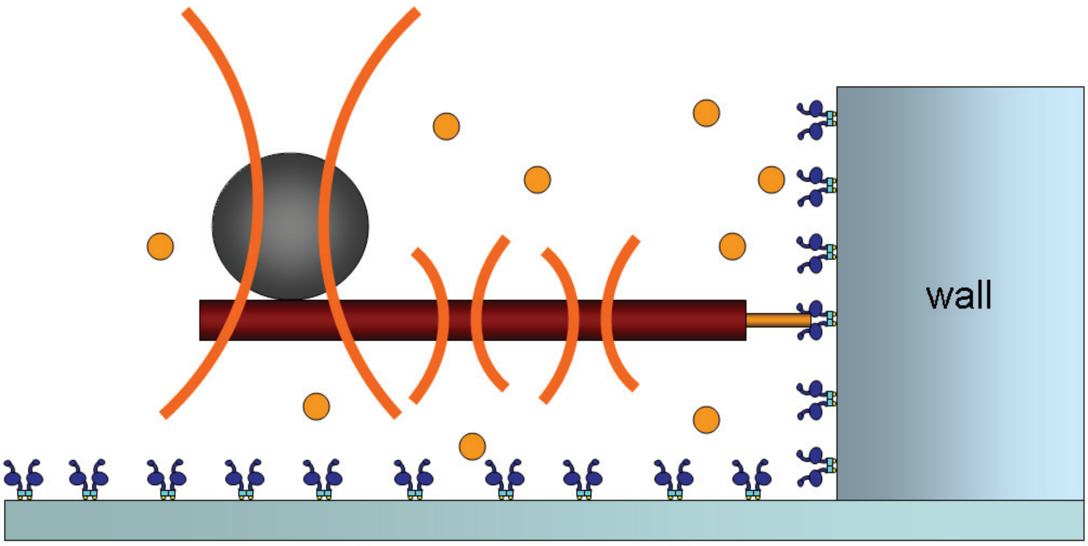
Microtubules are stiff, dynamic protein polymers that provide a mechanical framework (cytoskeleton) to living cells. Their growth and shrinkage generates pushing and pulling forces in the piconewton range that play a role in a variety of cellular motility processes. I will present *in vitro* experimental techniques involving optical tweezers that we use to study force generation by single microtubules *in vitro*. A bead is attached to a stiff microtubule-nucleating object (an axoneme), which is held with a time-shared "key-hole" optical trap in front of a microfabricated barrier (see schematic below). Growth of the microtubule results in motion of the bead that can be detected with nanometer resolution. Using this technique it is possible to study directly the molecular details of the microtubule growth process, as well as the forces that are generated at the dynamic microtubule end.



Specifically, I will show recent results on the interaction of dynamic microtubule ends with purified dynein motor proteins that are immobilized on our microfabricated barriers. In cells, pulling forces generated on microtubule ends by the minus-end directed motor protein dynein seem to play an important role in the positioning of the mitotic spindle, for example in budding yeast and *C elegans* embryos. Our results suggest that, on its own, dynein attached to a surface can capture dynamic microtubule plus ends and generate pulling forces on shrinking microtubules in an ATP-dependent way.