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Microenvironment of microtubules probed with fluorescence correlation spectroscopy.

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One aspect not completely understood regarding microtubule based transport is how motor proteins are able to move cargo through the cytoplasm when only a few are available to move each cargo and the force generated by each motor protein is small, 7 pN or less. This is surprising considering the high concentration of macromolecules and other proteins present in the cytoplasm through which cargo must move. To study the forces required to move organelle-sized particles through the cytoplasm, we have performed a series of measurements using magnetic tweezers. In these experiments, forces greater than 100 pN were required to move 0.3 µm magnetic beads in the cytoplasm. However, these experiments were performed on particles that had been injected into the cytoplasm and were not moving along microtubules. Transport of organelles at lower forces could be explained by spatial variation in the physical properties of the cytoplasm near microtubules. We have used fluorescent correlation spectroscopy (FCS) to compare the physical properties of the cytoplasm surrounding microtubules and elsewhere in the cell as a comparison. In PtK_2 cells expressing GFP-tubulin, fluorescently labeled 10 kD dextran was observed to have diffusion coefficients up to eight times higher near microtubules than elsewhere in the cell, demonstrating that microtubules are surrounded by a “high diffusion” zone. This spatial variation in the physical properties of the cytoplasm may facilitate transport of large cargo along microtubules.