Title
Two-photon single particle tracking: Investigation into chromatin movement.

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Authors
Ragan, T
Belmont, A
So, P
et al.

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Abstract

Recent research has indicated that the dynamics of chromatin movement inside of the nucleus may be much quicker than what was previously thought. For example, largescale chromatin structure reorganization induced by transcriptional activation has recently been observed. In order to study chromatin in vivo we are using a singleparticle tracking system that we have developed based around the two-photon microscope. The system is capable of tracking particles by either of two methods: The first method introduces aberrations into the optical system which break the axial symmetry of the point spread function and allows the 3D determination of the particle's position. The second method uses volumetric scanning to localize the particle. The system has an extended axial range of 100 microns and a frequency response of 15 Hz. To study the chromatin motion we have tagged regions of the chromosome by using lac repressor recognition of direct repeats of the lac operator with a GFP-repressor fusion protein. We present results on the motion of chromatin on the sub-second time scale and discuss the nature of the motion with respect to Brownian versus directed motion.