Transient anomalous subdiffusion of DNA-binding species in the nucleus

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Single-particle tracking experiments have measured the distribution of dwell times of various DNA-binding species — including CRISPR-Cas9, TetR, and LacI — diffusing in living cells. The observed truncated power law distribution has direct and indirect implications.

One direct implication is that the observed dwell time distribution is inconsistent with the Gaussian distribution of binding energies generally obtained from bioinformatics. Consideration of length scales of the nucleus and the measurement is essential to understanding the dwell time distribution.

Another direct implication is that a truncated power-law distribution of dwell times leads to transient anomalous subdiffusion, in which diffusion is anomalous, $\langle r^2 \rangle \propto t^{\alpha}$, $\alpha < 1$, at short times and normal, $\langle r^2 \rangle \propto t$, at long times, where $\langle r^2 \rangle$ is the mean-square displacement, t is time, and α is the anomalous diffusion exponent. The initial anomalous regime is of fundamental interest because it represents the search of the DNA-binding species for its target DNA sequence. Monte Carlo simulations are used to characterize the time-dependent diffusion coefficient D(t) and to relate the time dependence to the dwell time distribution. Time-dependent diffusion is described in terms of α, the limits D(0) and D(∞), and the crossover time between anomalous and normal diffusion. Detailed examination of the dependence of the subdiffusion parameters on the dwell time parameters is better suited to a paper than to a poster, but we give one example here. The trap concentration has a major effect on the anomalous diffusion exponent, but much of this effect is captured in D(0) and D(∞), which are easily calculated from the truncated power-law parameters, and are essential quantities to measure in experiments.

Some indirect consequences: (1). The simplest interpretation of the model is that the dwell times are actual binding times to DNA. One alternative is that the dwell times are the periods of one-dimensional diffusion on DNA in the standard combination of one-dimensional and three-dimensional search ("facilitated diffusion"), though the form of the dwell time distribution suggests a more complicated interpretation. (2). Non-target DNA sites have a significant effect on search kinetics; false positives from bioinformatic studies are potentially rate determining *in vivo*. (3). Overexpression of the DNA-binding species reduces anomalous subdiffusion because the deepest non-target binding sites are occupied and unavailable, a phenomenon well-known in the study of diffusion in catalyst pellets. (4). Both binding and obstruction affect diffusion. In the absence of a consensus model of chromatin geometry, obstruction effects ought to be characterized by experiment as well as by modeling. Suggested controls for obstruction are green fluorescent protein (GFP) as a calibration standard among laboratories and cell types, and the DNA-binding species itself with the binding site inactivated as unobtrusively as possible.

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