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Determining the Quantitative Dynamics of Nucleic Acids in Live Cells through Rics and iMSD Approaches

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Addressing the mobility and dynamics of DNA within live cells requires the characterization of individual particles in real time. Two such techniques enabling the quantification of the dynamics and means of motion of fluorescently labeled particles include the Raster Image Correlation Spectroscopy (RICS) and the image-based Mean Square Displacement (iMSD) approaches. In this study, the RICS and iMSD approaches were applied in order to elucidate the mobility and means of motion of DNA lipoplexes formed from varying sized DNA (20bp-5.5kbp). The RICS approach demonstrated that two species of mobility were present within the cytoplasm. Across the DNA fragment sizes, the slower species was not size dependent and were statistically the same. Whereas, faster moving particles which demonstrated a size dependent mobility. Nuclear localized DNA demonstrated to be significantly dynamic compared to other previously published work, ranging from 1.22 μm2/s (5.5kbp) to 3.67 μm2/s (21bp), consistent to nuclear proteins and RNA. The iMSD approach applied enabled the discrimination between different means of motion including random diffusion (RD), active transport (AT), confined diffusion (CD), anomalous subdiffusion (AS) and transient confinement (TC). Motion through RD and TC had demonstrated a size dependency across the DNA sizes assessed. Whereas, motion through AT, CD and AS did not demonstrate size dependency additionally these other means of motion had a higher mobility rate than that of RD.