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Probing Nucleosome Remodeling by Unzipping Single DNA Molecules

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At the core of eukaryotic chromatin is the nucleosome, which consists of 147 bp of DNA wrapped 1.65 turns around an octamer of histone proteins. Even this lowest level of genomic compaction presents a strong barrier to DNA-binding cellular factors that are required for essential processes such as transcription, DNA replication, recombination and repair. Chromatin remodeling enzymes use the energy of ATP hydrolysis to regulate accessibility of the genetic code by altering chromatin structure. While remodeling enzymes have been the subject of extensive research in recent years, their precise mechanism remains unclear. In order to probe the structure of individual nucleosomes and their remodeling, we assembled a histone octamer onto a DNA segment containing a strong nucleosome positioning sequence. As the DNA double helix was unzipped through the nucleosome using a feedback-enhanced optical trap, the presence of the nucleosome was detected as a series of dramatic increases in the tension in the DNA, followed by sudden tension reductions. Analysis of the unzipping force throughout the disruption accurately revealed the spatial location and fine structure of the nucleosome to near base pair precision. Using this approach, we investigate how remodeling enzymes may alter the location and structure of a nucleosome.