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Observing dynamics of chromatin fibers in Xenopus egg extracts by single DNA manipulation using a transverse magnetic tweezer setup JIE YAN, UIC Physics Department, DUNJA SKOKO, UIC Physics Department, JOHN MARKO, UIC Physics Department, TOM MARESCA, UC Berkeley, Department of Molecular and Cell Biology, REBECCA HEALD, UC Berkeley, Department of Molecular and Cell Biology — We have studied assembly of chromatin on single DNAs using Xenopus egg extracts and a specially designed magnetic tweezer setup which generates controlled force in the focal plane of the objective, allowing us to visualize and measure DNA extension under a wide range of constant tensions. We found, in the absence of ATP, interphase extracts assembled nucleosomes against DNA tensions of up to 3.5 piconewtons (pN). We observed force-induced disassembly and opening-closing fluctuations indicating our experiments were in mechanochemical equilibrium. We found that the ATP-depleted reaction can do mechanical work of 27 kcal/mol per nucleosome, providing a measurement of the free energy difference between core histone octamers on and off DNA. Addition of ATP leads to highly dynamic behavior: time courses show processive runs of assembly and disassembly of not observed in the -ATP case, with forces of 2 pN leading to nearly complete fiber disassembly. Our study shows that ATP hydrolysis plays a major role in nucleosome rearrangement and removal, and suggests that chromatin in vivo may be subject to continual assembly and disassembly.

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