INVITED LECTURE T9

Single-molecule studies of chromatin assembly in *Xenopus* egg extracts

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In this talk, we introduce our recent research of single-molecule studies of chromatins assembled in Xenopus egg extracts. In force-stretching experiments, we observes ~147 bp compaction steps at small forces and ~147 bp unfolding steps at large forces, suggesting nucleosome formation dominates the compaction of DNA in the Xenopus egg extract system. We found that, in the absence of ATP, interphase Xenopus extracts assembled nucleosomes against DNA tensions of up to 3.5 pN; force-induced disassembly and opening/closing fluctuations were observed; addition of ATP led to highly dynamic behavior time courses show processive runs of assembly and disassembly of not observed in the ATP-depleted case, with forces of 2 pN leading to nearly complete fiber disassembly. In addition, we have developed methods to image chromatins assembled in the extracts. Our methods allow us to image both the extended nucleosome-on-a-string structures and the more compact, higher order chromatin fibers under various reaction conditions. The combination of the chromatin manipulation and imaging techniques allow us to obtain both the chromatin assembly dynamics and chromatin conformational information.