ATP-induced helicase slippage reveals highly coordinated subunits

MICHELLE D. WANG, Cornell University

Helicases are vital enzymes that carry out strand separation of duplex nucleic acids during replication, repair and recombination. T7 helicase, a model hexameric motor, has been observed to use dTTP, but not ATP, to unwind dsDNA as it translocates along ssDNA. Whether and how different subunits of the helicase coordinate their chemo-mechanical activities and DNA binding during translocation is still under debate. Here we address this question using a single-molecule approach to monitor helicase unwinding. We found that T7 helicase does in fact unwind dsDNA in the presence of ATP and that the unwinding rate is even faster than that with dTTP. However, unwinding was repeatedly interrupted by sudden slippage events, ultimately preventing unwinding over a substantial distance. This behaviour was greatly reduced with the supplement of a small amount of dTTP. These findings presented an opportunity to use nucleotide mixtures to investigate helicase subunit coordination. Our results support a model where nearly all subunits coordinate their chemo-mechanical activities and DNA binding. Such subunit coordination may be general to many ring-shaped helicases and reveals a potential mechanism for regulation of DNA unwinding during replication.