Basic Research

Single-molecule Studies Reveal a Traveling Wave-like Model for Homologous Recombination

Homologous recombination (HR) is a major pathway for repair of double-stranded DNA break. RecA polymerizes along the single-stranded DNA at the end of a broken DNA to form a presynaptic filament, which is a right-handed helix with ~6 monomers per turn and 3 bases per monomer. The presynaptic filament catalyzes the search for homologous sequences and promotes the strand exchange with a donor DNA. HR has been known for decades, but the underlying molecular mechanism remained unclear. By using single molecular magnetic tweezers (MT) and fluorescent resonant energy transfer (FRET), the lab led by Prof. LI Ming at the Institute of Physics (IOP), Chinese Academy of Sciences (CAS), managed to catch the transient states during HR and build a traveling model for RecA-mediated HR.

In an effort to solve the challenge of catching the transient states, the researchers realized that the

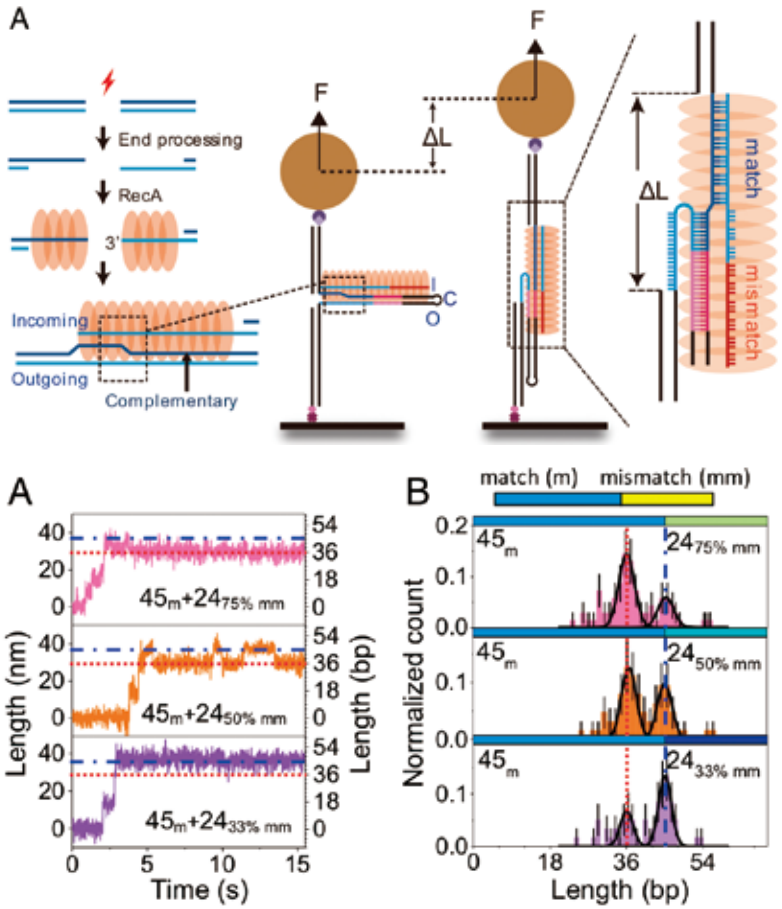

Figure 1: MT assay of the strand exchange. (Image by Institute of Physics)

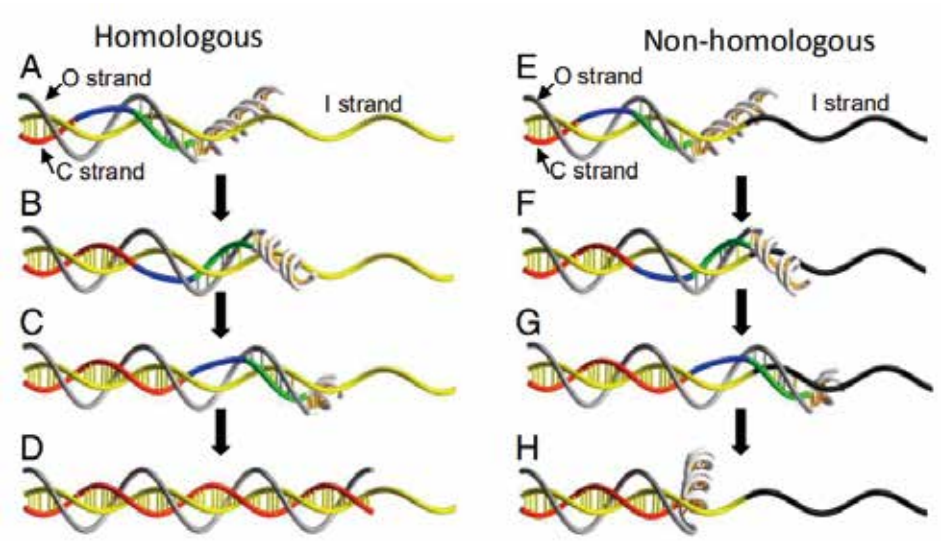

Figure 2: Stepwise progression in HR. (Image by Institute of Physics)

question of how the strand exchange progresses is actually equivalent to how it is blocked. They therefore designed a series of mismatch-containing donor DNAs to assess the dynamics of HR at the single-molecule level. They found that even though 3 base pairs (bp) is still the basic unit as previously reported, both the homology checking and the strand exchange may proceed in multiple steps at a time, resulting in ~9 bp large steps on average. More interestingly, the strand exchange is blocked remotely by the mismatched segment, terminating at positions ~9 bp before the match-mismatch joint.

The results suggest that the homology checking and the strand exchange are separated in space, with the strand exchange lagging behind the homology checking by ~9 bp. In order to explain their observations, the researchers made a hypothesis that there exists a transitional segment between the homology-checking region and the post strand-exchanged region. Accordingly, they built a model of strand exchange: the strand exchange progresses like a traveling wave in which the donor DNA is incorporated successively into the ssDNA-RecA filament to check homology in ~9-bp steps in the frontier, followed by a hypothetical transitional segment and then the post strand-exchanged duplex (Figure 2).

This study entitled "Mismatch sensing by nucleofilament deciphers mechanism of RecA-mediated homologous recombination" was published in *PNAS*.

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