P-1.180

Resolving Stable G-quadruplexes: Pif1 Dynamics in Dsdna and Kras Promoter G4 Stability

Jessica Valle Orero [1], Jean-Francois Allemand [2], Vincent Croquette [2], Jean-Baptiste Boule [3]

[1] Ecole Normale Superieure / The Amercian University of Paris, PARIS, France

[2] Ecole Normale Superieure, Paris, France

[3] Muséum national d'Histoire naturelle, Paris, France

G-quadruplexes (G4) are stable DNA and RNA structures formed by guanine-rich sequences, found in key genomic regions such as telomeres, promoters, and replication origins. These structures can impede replication and transcription, requiring specialized helicases like Pif1 to resolve them. While Pif1's role in resolving G4s in single-stranded DNA is well studied, its activity in double-stranded DNA (dsDNA) remains less understood.

We developed dsDNA assays to induce highly stable G4 structures (c-Myc G4, lifetime >1 hour) and observed Pif1's real-time action in resolving them. Our results show that Pif1 reduces G4 lifetime from hours to seconds, resuming translocation without transitioning from monomer to dimer, contrary to previous reports. Additionally, we observed strand-switching behavior in the presence of G4s.

We also investigate G4 structures in the KRAS promoter, a gene frequently mutated in cancer. These structures may regulate KRAS transcription, making them potential therapeutic targets. Using a novel single-molecule approach, we analyze their stability and interactions with regulatory proteins, providing insights critical for developing strategies to disrupt KRAS transcription in cancer.