Robert M Brosh et al., Transcriptomics 2017, 5:2 (Suppl)
DOI: 10.4172/2329-8936-C1-012

Analyses of patient-derived missense mutations in Fanconi anemia group J (FANCJ) DNA helicase

Robert M Brosh, Sanjay Kumar Bharti, Irfan Khan, Joshua A and **Sommers**
National Institute on Aging, NIH, USA

Statement of the Problem: Fanconi anemia (FA) is a rare genetic DNA repair disorder characterized by progressive bone marrow failure, congenital abnormalities, and cancer. Of the 21 genes linked to FA, the FA Group J (FANCJ) gene is unique that it encodes an ATP-dependent DNA helicase. Mutations in *FANCJ* are not only genetically linked to FA, but also associated with breast and ovarian cancer. Consistent with its known role in homologous recombination (HR) repair, FANCJ-/- cells are sensitive to DNA interstrand cross-linking (ICL) agents and are also hypersensitive to agents that induce replication stress.

Methodology & Theoretical Orientation: We characterized two FA patient-derived *FANCJ* mutations, *R707C* and , which reside in the conserved helicase core domain. Genetic and biochemical analyses were performed to delineate the molecular defects underlying the genetic disease.

Findings: *FANCJ-R707C* retained partial (~30%) helicase activity, whereas *FANCJ-* was nearly completely inactive. Single-turnover kinetic assays, ATPase measurements, and DNA binding determinations confirmed the differential effects of *FANCJ* missense mutations on helicase activity. Expression of either *FANCJ-R707C* or *FANCJ-* in *FANCJ-/-* cells completely failed to rescue cisplatin sensitivity. In striking contrast, expression of FANCJ-*R707C* in *FANCJ-/-* cells restored resistance to the DNA polymerase inhibitor aphidicolin, whereas FANCJ- completely failed. Single-molecule replication tract analysis confirmed that *FANCJ-R707C*, but not FANCJ-, restored fork rates after cellular exposure to aphidicolin. Thus, a quantitatively lower threshold of *FANCJ* catalytic activity is required for the aphidicolin-induced replication stress response compared to cisplatin-induced damage.

Conclusion & Significance: The catalytic requirement of *FANCJ* to reconstruct broken replication forks after ICL-induced damage is distinct from that required to remodel stalled replication forks. These findings provide new insight to FANCJ's role in DNA repair and molecular phenotypes of clinically relevant *FANCJ* missense mutations that are relevant to human disease and cancer.

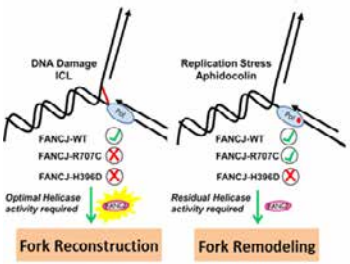

Fig. 1. Divergent roles of FANCJ in ICL repair and response to replication stress dictated by catalytic threshold requirement.

Biography

Robert M Brosh has his expertise in DNA Repair and Genome Stability Maintenance. He leads a research group at the National Institute on Aging, NIH that is focused on characterizing the roles of clinically relevant human DNA helicases in cellular nucleic acid metabolism. This work has yielded insights into how DNA repair helicases promote phenotypes consistent with healthy aging and cancer resistance.

broshr@mail.nih.gov

Notes: