FALL 2023

Biochemistry and Molecular Biology Brown Bag Series

Resha Shrestha

BMS Ph.D. Student

"Human Genome instability at $(CAG)_{102}$ Microsatellite DNA Repeats"

Tuesday, October 31, 2023

11:00 AM

135 Oelman Hall

Lab: Michael Leffak, Ph.D.

Boonshoft School of Medicine
WRIGHT STATE UNIVERSITY

WRIGHT STATE UNIVERSITY

https://science-math.wright.edu/biochemistry-and-molecular-biology

Abstract:

Human Genome instability at $(CAG)_{102}$ Microsatellite DNA Repeats

Microsatellites are tandem repeats of short nucleotide sequences that are inherently unstable. These repeats can form non-Watson-Crick structures that can pose obstacles to DNA replication leading to double strand breaks (DSBs). When unrepaired, these breaks can threaten genomic integrity and cause various neurological diseases and cancers. These replication-induced breaks can be repaired by a highly mutagenic mechanism known as break induced replication (BIR). Here, we focus on CAG trinucleotide repeats capable of forming hairpin structures which stall the replication fork, eventually causing its collapse. Expansions in CAG repeats have been implicated in Huntington's disease and Myotonic Dystrophy. To study instability caused by this repeat, we are using an engineered HeLa cell line with dual fluorescent reporter constructs harboring a repeat of 102 CAG units in the lagging strand template adjacent to a c-myc origin of replication. This allows us to use flow cytometry, inverse PCR, and DNA sequencing to study the effects of these repeats at the single DNA molecule level. We find that the repair of these replication-induced breaks leads to 1000x elevated levels of DNA mutagenesis including insertions, deletions, base substitutions and the formation of extrachromosomal circular DNAs (eccDNAs).