UNIVERSITY of GUELPH

COLLEGE of BIOLOGICAL SCIENCE

DEPARTMENT OF MOLECULAR AND CELLULAR BIOLOGY

Announcement:

All interested members of the university community are invited to attend the Final Oral Examination for the degree of ***Master of Science*** *of*

SAMANTA PLADWIG

on Tuesday, August 25, 2020 at 9:30 a.m. (online)

Thesis Title: **Characterization of human BRCA2 truncations on Rad51 levels and homology directed repair in mammalian cells**

Examination Committee:

Dr. George van der Merwe, Dept. of Molecular and Cellular Biology (Exam Chair)
Dr. Mark Baker, Dept. of Molecular and Cellular Biology
Dr. Richard Mosser, Dept. of Molecular and Cellular Biology
Dr. Wei Zhang, Dept. of Molecular and Cellular Biology

Advisory Committee:
Dr. Mark Baker (Advisor)
Dr. Richard Mosser
Dr. Ray Lu

Abstract: Two keystone proteins in the homology-directed repair (HDR) pathway for the restoration of double stranded DNA breaks (DSBs) are BRCA2 and Rad51, which interact through eight highly conserved motifs known as BRC repeats located within the centre of BRCA2. BRCA2 recruits Rad51 to processed single stranded DNA ends at the site of the break to stimulate the formation of nucleoprotein filaments which search for and invade homologous sequences, utilizing them as a template to repair the break with high fidelity. Germline mutations in BRCA2 increase the risk of developing early onset cancer, such as pathogenic variant 6174delT which truncates BRCA2 within the 7th BRC repeat and eliminates key functional domains in the C-terminus, including DNA binding domains and nuclear localization signals. Mouse hybridoma cells (igm482) expressing 2XMBP-tagged human BRCA2 proteins truncated after the 8th, 6th, 4th and just before the 1st repeat, were used to investigate how the number of BRC repeats present in a truncated BRCA2 protein impacts Rad51 protein levels and efficiency of HDR. Cell growth, DNA damage tolerance and p53 protein levels were also characterized, with each BRCA2 truncation variant producing a unique cellular response. The variable effects observed by changing the number of BRC repeats in the context of a missing C-terminal domain underscores the complexity of BRCA2, both in its role in promoting HDR and how dysfunctional proteins lead to tumorigenesis. This work highlights the importance of elucidating how dysfunctional proteins effect cellular homeostasis in order to identify effective treatments, as well as new therapeutic targets.

Curriculum Vitae: Samanta completed her Bachelor of Science (Hons.) at the University of Guelph in the spring of 2017 and then began her MSc later that fall in the lab of Dr. Mark Baker.

Awards: Ontario Graduate Scholarship, May 2018

Publications: Magwood, A.C., Mundia, M.M., **Pladwig, S.M.**, Mosser, D.D., and Baker, M.D. (2020). The dichotomous effects of caffeine on homologous recombination in mammalian cells. DNA Repair 88, 102805.