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

IQRA RAZZAQ

on Wednesday, February 3, 2021 at 1:30 p.m. (online)

Thesis Title: Analyzing the roles of the essential gene TRA1 and the novel extracellular vesicle EVP1 in regulating morphogenesis and the antifungal drug resistance response in the opportunistic fungal pathogen Candida albicans

Examination Committee:
Dr. Mike Emes, Dept. of Molecular and Cellular Biology (Exam Chair)
Dr. Rebecca Shapiro, Dept. of Molecular and Cellular Biology
Dr. Jennifer Geddes-McAlister, Dept. of Molecular and Cellular Biology
Dr. Joseph Yankulov, Dept. of Molecular and Cellular Biology

Advisory Committee:
Dr. R. Shapiro (Advisor)
Dr. C. Khursigara
Dr. J. Geddes-McAlister

Abstract: Fungal pathogens, which have historically been understudied, are important disease-causing agents with global significance. Candida albicans is an opportunistic fungal pathogen and the leading cause of candidiasis, a systemic bloodstream infection with 40% mortality. Dynamic morphogenic plasticity allows C. albicans to evade host immune responses and rapidly adapt to new environments. Additionally, C. albicans possesses a diverse arsenal of adaptive mechanisms, which confer resistance to all three classifications of antifungal drugs (azoles, polyenes, and echinocandins), emphasizing the importance of exploring new targets for the development of novel antifungals. The role of the essential gene TRA1 in C. albicans has not previously been studied in literature. We used CRISPR strategies to introduce three arginine to glutamine point mutations into TRA1 (R3471Q, R3472Q, and R3538Q), which have previously been shown to confer antifungal sensitivities when introduced into the non-pathogenic yeast model S. cerevisiae, generating a tralQ3 mutant. Phenotypic profiling of tralQ3 revealed that the mutant is highly azole resistant to fluconazole and miconazole, while subsequently demonstrating hypersensitivity to cell wall stressors caspofungin and calcofluor white, demonstrating an example of collateral sensitivities within fungi, a phenomenon previously observed only in bacteria and cancer cells. RNA-seq analysis of tralQ3 revealed 289 differentially expressed genes between tralQ3 and the WT, of which 188 were downregulated in the mutant and 101 genes were upregulated. From this data, we identified and further explored the gene Orf19.6741, which has not been studied in literature, and created a novel ΔOrf19.6741 knockout mutant (Δevp1), which demonstrated azole hypersensitivity. Both tralQ3 and Δevp1 demonstrated impaired filamentation and biofilm formation, antifungal sensitivity, and reduced macrophage evasion, indicating their potential involvement in multiple stress response pathways. Together, this work demonstrates the novel cellular roles of two previously uncharacterized genes in C. albicans.

Curriculum Vitae: Iqra completed her Bachelor of Science (Hons.) at the University of Guelph in the summer of 2018, and then began her MSc in the lab of Dr. Rebecca Shapiro in the fall of the same year.