

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 **Doctor of Philosophy** of

BARRET FOSTER

on Wednesday, December 4th, 2024 at 9:30a.m. (SSC 1504)

Thesis Title: Analysis of fermentation capabilities, trehalose metabolism, quantitative proteomics, and hybridisation of Norwegian kveik brewing yeast

Examination Committee:

Dr. Nina Jones, Molecular and Cellular Biology (Exam Chair) Dr. George van der Merwe, Dept. of Molecular and Cellular Biology Dr. Krassimir Yankulov, Dept. of Molecular and Cellular Biology Dr. Jennifer Geddes-McAlister, Dept. of Molecular and Cellular Biology Dr. Debbie Inglis, Biological Sciences Department, Brock University (External Examiner)

Advisory Committee:

Dr. George van der Merwe (Adv) Dr. Steffen Graether

- Dr. Rebecca Shapiro
- Dr. Krassimir Yankulov

Abstract: Kveik, a group of farmhouse *Saccharomyces cerevisiae* ale yeast from Western Norway, have recently gained immense popularity. Despite their popularity, kveik remain a mystery; their fermentation capabilities have not been well characterized, the exact mechanisms of their stress adaptations have not been elucidated, and their potential to create novel hybrid yeast is unexplored. In this thesis, I characterized the fermentation profiles of kveik strains across a range of temperatures to analyze fermentation characteristics, carbon metabolism and metabolite profiles in lab-scale wort fermentations. I report that most kveik are capable of fermenting efficiently at a wider range of temperatures and at significantly warmer temperatures than most industrial ale strains. Viability analysis revealed that the improved warm fermentation performance is due to kveik strains being able to survive at much higher temperatures, likely as a result of their ability to produce and maintain significant concentrations of intracellular trehalose. I identified deficient neutral trehalase activity in kveik as the likely mechanism for improved trehalose accumulation. To identify additional stress adaptations, I performed label-free quantitative proteomic analysis of kveik strains undergoing a prolonged thermal stress to analyze how the proteomes of these strains respond to severe heat and how their response differs from less tolerant strains. From this total proteome analysis, I identified several mechanisms in kveik which likely contribute to their stress tolerance. These include modulations to the central carbon metabolism, a significantly higher abundance of the ROS-detoxifying catalase Ctt1, an enrichment of ergosterol biosynthetic enzymes, and potentially enhanced mitochondrial function relating to ATP synthesis. Additionally, I report the first instance of successfully using Norwegian kveik as breeding stock in spore-to-spore mating trials with industrial Beer 2 ale strains to create novel kveik hybrid ale yeasts with strong fermentation capabilities and stress tolerant characteristics with the potential for immediate application in the brewing industry.

This research has important implications for both the brewing industry and the broader understanding of stress tolerance in yeast. Further investigation of the stress adaptations of Norwegian kveik and implementation of hybridization trials will allow us to maximize the contribution of kveik yeasts to fermentation industries.

Curriculum Vitae: Barret obtained his Bachelor of Science in Molecular Biology and Genetics with a minor in Biotechnology at the University of Guelph in 2018. In the fall of 2018, he entered into the MSc. program under the supervision of Dr. George van der Merwe. In the winter of 2020, he transferred into the PhD program.

Publications: Foster, B., Tyrawa, C., Ozsahin, E., Lubberts, M., Krogerus, K., Preiss, R., & van der Merwe, G. (2022). Kveik Brewing Yeasts Demonstrate Wide Flexibility in Beer Fermentation Temperature Tolerance and Exhibit Enhanced Trehalose Accumulation. *Frontiers in microbiology, 13*, 747546. https://doi.org/10.3389/fmicb.2022.747546

Rowlands, H., Shaban, K., Foster, B., Proteau, Y., & Yankulov, K. (2019). Histone chaperones and the Rrm3p helicase regulate flocculation in S. cerevisiae. *Epigenetics & chromatin*, *12*(1), 56. https://doi.org/10.1186/s13072-019-0303-8

Preiss, R., Fletcher, E., Garshol, L. M., Foster, B., Ozsahin, E., Lubberts, M., van der Merwe, G., & Krogerus, K. (2024). European farmhouse brewing yeasts form a distinct genetic group. *Applied microbiology and biotechnology*, *108*(1), 430. https://doi.org/10.1007/s00253-024-13267-3

Rowlands, H., Shaban, K., Cheng, A., Foster, B., & Yankulov, K. (2019). Dysfunctional CAF-I reveals its role in cell cycle progression and differential regulation of gene silencing. Cell cycle (*Georgetown*, *Tex.*), *18*(22), 3223–3236. https://doi.org/10.1080/15384101.2019.1673100