

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

ALEXANDER NOVODVORSKI

On Friday, December 15, 2023 at 1:30 p.m. (SSC 2315)

Thesis Title: Characterization of putative probiotic *Bacillus* spp. isolates that target *Clostridium perfringens* for controlling chicken necrotic enteritis

Examination Committee:

Dr. Marc Coppolino, Dept. of Molecular and Cellular Biology (Exam Chair)Dr. Stephen Seah, Dept. of Molecular and Cellular BiologyDr. Dr. Jennifer Geddes-McAlister, Dept. of Molecular and Cellular BiologyDr. Steffen Graether, Dept. of Molecular and Cellular Biology

Advisory Committee:

Dr. Stephen Seah (Co-Advisor) Dr. Joshua Gong (Co-Advisor) Dr. Jennifer Geddes-McAlister Dr. Jason Carere

Abstract: *Bacillus amyloliquefaciens* HG88 and *B. subtilis* CG46, were isolated from antibiotic-free chicken intestine samples and their cell free supernatants (CFS) were found to inhibit the growth of *Clostridium perfringens*, the causative agent of necrotic enteritis in chickens. Their inhibitory compounds were determined to be proteinaceous due to their susceptibility to protease digestion. Separation of proteins from the CFS of the two *Bacillus* strains followed by peptide mass fingerprinting enabled the identification of putative antimicrobial peptides (AMP) with export signals for secretion from the cell. The peptide from *B. amyloliquefaciens* HG88, IP_{HG88}, is homologous to bacterial SH3 domains that were known to bind to the peptide portion of peptidoglycan. The gene encoding this peptide was cloned and the peptide was purified from recombinant *Escherichia coli* as an N-terminal His-tagged protein. His-tagged IP_{HG88} inhibited growth of *C. perfringens* CP1 with a minimum inhibitory concentration (MIC) of ~4.96 µg/mL. Microscopic examination revealed morphological defects in the *C. perfringens* cells characterized by elongated or filamentous cells. The second putative inhibitory peptide, from *B. subtilis* CG46, is homologous to a previously characterized *B. subtilis* AMP, named LCI, that has broad spectrum activity against many bacterial species but had not previously been shown to be inhibitory towards *C. perfringens*.

Curriculum Vitae: Alexander completed his Bachelor of Science in Biochemistry at the University of Waterloo in 2021. He began his Master of Science program in Molecular and Cellular Biology at the University of Guelph in Dr. Seah's lab that same year.