**The Dark Side of Cyanobacteria: Expanding Our Understanding of Non-photosynthetic Cyanobacteria in Freshwater Systems**

Harshina Brijlall1; Anjali Krishna1, Monica Emelko2, and Kirsten Müller1

*1 Department of Biology, University of Waterloo, Waterloo, Ontario*

*2 Department of Civil and Environmental Engineering, University of Waterloo, Waterloo, Ontario*

Genomic sequencing has expanded the phylum Cyanobacteria to include Non-photosynthetic Cyanobacteria (NCY). The NCY lack photosynthetic mechanisms and thrive in darker environments. This group is also hypothesized to be cyanotoxin producers of β-Methylamino-L-alanine (neurotoxin). The objective of this study is to explore the biogeography and seasonal distribution of NCY in freshwater systems. The Turkey Lakes Watershed (TLW), in Ontario, is a model site to observe the influence of autumnal and vernal shifts on cyanobacterial community dynamics in a changing climate. Using the 16S rRNA gene V4 region, amplicon sequencing was performed, and taxonomy was assigned with the SILVA v138 classifier. The NCY taxa observed for the summer months in TLW from 2018 to 2022 were Gastranaerophilales, Obscuribacterales, Vampirovibrionales, and Caenarcaniphilales. Samples collected in Summer 2022 and Fall 2023 reported 5 ASVs with a frequency of 160 reads (total) identified as NCY. Integrated water column samples from Winter 2023 reported a diversity of NCY, implying that ice cover may play a role in NCY community. In addition, NCY distribution were observed across five ecozones in Canada. The classification of NCY within Cyanobacteria is highly debated and therefore investigating NCY distribution is vital to microbial ecology.