## Identifying the effects of N-glycan differences between *Nicotiana benthamiana* and *Pichia* pastoris on recombinant enzymes

Nicole Fraser<sup>1</sup>, Nardo Nava<sup>2</sup>, and David Rose<sup>3</sup>

<sup>1</sup>University of Waterloo, Waterloo, Ontario, Canada, <u>n3fraser@uwaterloo.ca</u>

<sup>2</sup> nenavaro@uwaterloo.ca

<sup>3</sup> david.rose@uwaterloo.ca

Glycosylation is one of the most abundant forms of post-translational modifications in proteins<sup>1</sup>. Different organisms produce different N-glycans. As determined in literature, there are predicted severe differences in glycans produced in plants versus yeast. N-glycans can be categorized into three main products: high-mannose, complex, and hybrid. Plants produce complex glycans whereas yeast produce high-mannose glycans<sup>2</sup>. These structures differ by their monosaccharide composition and their linkages which could alter the behaviour and structure of the enzyme. *N.benthamiana* (plant) and *P.pastoris* (yeast) are two organisms that are predominant in recombinant protein expression for their ability to express large proteins with post-translational modifications. The focus of this project is to define the effects of different N-glycans on enzymes through quantifying differences in activity, stability and potentially – structure. Current work has been focusing on successfully expressing enzymes within both systems and quantifying the differences in glycosylation to elucidate these unknown mechanisms.

## **References**

[1] Roth, Z. Yehezkel, G. & Khalaila, I. (2012). International Journal of Carbohydrate Chemistry, 2012, 1-10.

[2] Strasser, R. (2016). Glycobiology, 2016, 926-939.