Abstract

This bachelor thesis is part of a project aiming for the crystallization of trehalase domain of neutral trehalase Nth1 from *Saccharomyces cerevisiae*. The main goal of this thesis is to optimize the purification protocol of two different constructs of yeast Nth1 and determine the optimal buffer for crystallography of these constructs using the differential scanning fluorimetry DSF.

Trehalases are important, highly conserved enzymes found in many organisms. These enzymes belong to the class of hydrolases, subgroup of glycosidases and their common feature is hydrolytic cleavage of trehalose molecule into two glucose subunits. Trehalose is a naturally occurring non-reducing disaccharide which serve in yeast cells as storage carbohydrate and energy and stress metabolite. Trehalose and its hydrolysis is very important in the life cycle of the insect, as it is present as the main sugar component of insect hemolymph, so some trehalase inhibitors could be used as insecticides. By this time, however, only one structure of trehalase from a prokaryotic organism *Escherichia coli* was solved. For this reason it is very important to solve the structure of trehalase from a eukaryotic organism.

Keywords:

Nth1, expresion, purification, DSF

Key subjects:

Protein biochemistry, biophysical chemistry