Presenter: Ruiyue Tan

Authors: Ruiyue Tan, Ethan Walker, John Bettinger, Michael Meadow, Sina Ghaemmaghami

Title: The effect of the ribosome on the thermodynamic stability of nascent proteins

Abstract

Proteins exist in a dynamic equilibrium between folded and unfolded conformations. The free energy difference between these two states (ΔGfolding or 'thermodynamic folding stability') establishes the fraction of the protein population that is in a folded conformation at equilibrium. Folding stabilities can impact the tendency of proteins to misfold, aggregate, oxidize and undergo degradation. They also play a role in regulating protein function in many biological processes and their alteration has been associated with a number of pathological conditions. Historically, folding stabilities have been investigated for full-length proteins in purified solutions. However, within a cell, proteins are known to fold during the course of translation. As they fold, the C-termini of nascent polypeptides are restricted in the exit tunnel of the ribosome. How this close proximity to the ribosome impacts the stability and folding of nascent polypeptides remains largely unknown. Recently, our laboratory has developed a mass spectrometry-based methodology that is capable of measuring protein ΔGfolding values by quantifying rates of oxidation for methionines buried in the protein core (SPROX). Importantly, SPROX enables the measurement of ΔGfolding in complex and unpurified protein mixtures. Using this technique, we investigated the impact of the ribosome on the folding stabilities of nascent polypeptides in in vitro translation assays. We measured folding stability of three proteins: DHFR, CheY and Pol IV. The results showed that the folding stability of nascent polypeptides destabilized when proximal to ribosome surface. This destabilization may help with protein folding into compact conformation and facilitate co-translational enzyme modification.