Determination of Optimal Metal Conditions for Adaptation in a Type 1E CRISPR System

DeBell, Lily (School: Baltimore Polytechnic Institute)

CRISPR-Cas systems are the adaptive immune systems of bacteria and archaea. CRISPR systems allow cells to develop immunological memory by storing information from foreign DNA in the bacterial or archaeal genome itself. Adaptation, the stage of CRISPR immunity during which foreign DNA is incorporated into the prokaryotic genome, is an essential but poorly understood aspect of CRISPR-Cas immunity. Cas1 is a metal-dependent endonuclease required for recognition, processing, and integration of foreign DNA during adaptation. While it is has been demonstrated that Cas1 requires the presence of a divalent metal cation for its nuclease and integrase activity, it is unclear which metal is preferred by Cas1 for robust nuclease and integration activity. I determined optimal metal conditions for Cas1-Cas2 facilitated integration of DNA into a CRISPR array in an in vitro E. coli CRISPR system. In the presence of Mn2+, integration occurred more quickly and robustly than in the presence of Mg2+. Future work includes determining preferred metal ligand for Cas1 based on active site structure and which metal cation best supports the processing and trimming functions of the nuclease during adaptation.

Awards Won: