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University of Ottawa

Ottawa

Corresponding Faculty Member: Adam Rudner (arudner@uottawa.ca)

A novel +2 translational frameshift in tail assembly chaperones of EA cluster bacteriophages.

Enzhe Galimova, Laura Giles, Elizabeth C Williams, Adam D Rudner

Translational frameshifting is a mechanism that allows cells to produce two versions of a specific protein, a N-terminal short form, and a longer form created by the ribosome slipping either forward or backwards during the mRNA translation process, causing a frameshift that extends the protein sequence. Frameshifting in bacteriophage tail assembly chaperones (TAC) has been previously studied, however, there is no experimental evidence of translational frameshifting of the TAC proteins in EA cluster bacteriophages. In this study, we use a polyclonal antibody that recognizes the long form of the TAC to demonstrate that Winzigespinne (an EA1 phage) and Quartz (an EA10 phage) produce the frameshifted TAC in vivo during an infection of their *MIcrobacterium foliorum* host. To determine the precise slippery sequence we performed mass spectrometry on frameshifted protein purified from E. coli and have identified a novel consensus slippery sequence, GGGXGA, which leads to a +2 frameshift. Our work will lead to changes in the annotation of many EA cluster bacteriophages, and is the first experimental evidence of a regulated +2 ribosomal frameshift. Translational frameshifting also occurs in several animal viruses, including HIV and Influenza, and this frameshifting also regulates the ratios of viral proteins. Our work is broadly applicable to understanding and exploiting the fundamental mechanisms that trigger ribosomal slippage.