2017

express recombinant Tb12990 protein from E. coli for DNA binding analysis

Alex Lotozynski
Cleveland State University

Express recombinant Tb12990 protein from E. coli for DNA binding analysis

College of Sciences and Health Professions

Student Researcher: Alexander Lotozynski

Faculty Advisor: Bibo Li

Abstract

Trypanosoma brucei is a parasitic protozoan species, causing sleeping sickness in humans and nagana in livestock. Its vector, the tsetse fly, feeds on an infected host and passes the parasites into the bloodstream of other mammalian hosts. The unique challenge for elimination is its complex process of antigenic variation, where the parasite regularly switches its major surface antigen, VSG. When the parasite enters the mammalian host, the host responds by making a corresponding antibody against its major surface antigen, VSG. However, although most parasites are eliminated, a small population can escape due to their altered VSG coat. The expression sites for VSGs are near the telomeres of T. brucei. Our lab has found that telomere proteins, including TbRAP1, TbTRF, and TbTIF2, suppress this VSG switching. TbRAP1 also regulates the VSG silencing. Using TbRAP1 as bait in Yeast Two-Hybrid screens, we identified Tb12990 as a potential TbRAP1-interacting factor. Sequence analysis suggests that Tb12990 might be a homologue of the vertebrate telomere protein TbTPP1. In order to examine whether Tb12990 has any telomere DNA binding activities as its homologues, we intend to express its recombinant protein in E. coli. We have cloned Tb12990 in pGEX-4T- 2 and pET-15b. We will next purify the Tb12990 from E. coli.