M & I
Microbiology
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DISSERTATION SEMINAR

Heather A. Vincent

"Regulation of mRNA Translation by Human Cytomegalovirus pTRS1"

Thursday, November 30, 2017

3:00 p.m.

6004 Marsico Hall

Dissertation Advisor: Dr. Nathaniel Moorman

Presented in partial fulfillment of the requirements for the degree of Doctor of Philosophy

ABSTRACT

Heather Ashley Vincent: “Regulation of mRNA Translation by Human Cytomegalovirus pTRS1”
(Under the direction of Nathaniel Moorman)

Human cytomegalovirus (HCMV) is a major public health burden. Acute infection during pregnancy can lead to congenital birth defects, and reactivation of a latent infection in immune compromised individuals can cause significant morbidity and mortality. HCMV does not encode its own ribosomes, and is therefore completely reliant on the host translation machinery for viral protein synthesis. HCMV also does not induce host translational shutoff upon infection, thus viral mRNAs must compete with cellular mRNAs to efficiently translate viral proteins. The HCMV protein TRS1 (pTRS1) plays an integral role in translation regulation during HCMV replication by antagonizing the antiviral kinase PKR. Activated PKR phosphorylates eIF2α, which causes an overall inhibition of protein synthesis that inhibits HCMV replication. pTRS1 also increases overall levels of protein synthesis and enhances the translation of reporter mRNAs in a PKR-independent manner, showing that pTRS1 regulates mRNA translation through multiple mechanisms. In this dissertation I sought to define the mechanisms used by pTRS1 to stimulate translation. In chapter 1 I show that pTRS1 inhibits PKR activation by binding to PKR and inhibiting PKR kinase activity. pTRS1 binds PKR residues that are conserved across eIF2α kinases, suggesting that pTRS1 can antagonize multiple eIF2α kinases. In chapter 2 I show that pTRS1 stimulates cap-independent translation. pTRS1 enhances the activity of both host and viral internal ribosome entry sites (IRESs) and stimulates translation of a circular mRNA reporter. These pTRS1 functions were independent of its ability to antagonize PKR, but dependent on its ability to bind double-stranded RNA. To understand how pTRS1 stimulates translation, in chapter 4 I identify ribosome-associated, cellular proteins that bind pTRS1. I found that pTRS1 interacts with active protein phosphatase 1 (PP1) catalytic subunits. Rather than affect PP1 catalytic activity, pTRS1 changes the complement of proteins that interact with the PP1 alpha catalytic subunit, possibly to regulate PP1 substrate specificity. Together these data further characterize the mechanisms used by pTRS1 to regulate mRNA translation and reveal how pTRS1 may contribute to the efficient translation of viral mRNAs during HCMV infection.