Mercer University Electronic Thesis/Dissertation Submission Form

☐ Thesis  □ Dissertation  □ Restricted
If Restricted, additional form required

Date: 01/20/2017  Due Date: 7/1/2017

Name: Ellie Kopfeind  Student ID: 104694368

College/School: School of Medicine  Campus Location: Savannah

Department/Program: Biomedical Science

Director of Graduate Program: Dr. Richard McCann

Degree Granted: Master of Science  Graduation Date: May 2017

Title of Thesis or Dissertation: Intermolecular complementation between pORF64 mutant monomers as a mechanism to study Varicella Zoster Portal Domains

Attachments:
☐ Dissertation Title Page
☐ Abstract Page
☒ Signed Approvals Page
INTERMOLECULAR COMPLEMENTATION BETWEEN pORF54 MUTANT MONOMERS AS A MECHANISM TO STUDY VARICELLA-ZOSTER PORTAL DOMAINS

by

ELLIE M. KORNFEIND

A Thesis Submitted to the Faculty of the School of Medicine at Mercer University in Partial Fulfillment of the Requirements for the Degree

MASTERS IN BIOMEDICAL SCIENCES

Savannah, GA
ABSTRACT

ELLIE M. KORNFEIND

INTERMOLECULAR COMPLEMENTATION BETWEEN pORF54 MUTANT MONOMERS AS A MECHANISM TO STUDY VARICELLA-ZOSTER PORTAL DOMAINS

Under the direction of ROBERT VISALLI, Ph.D.

There are nine human herpes viruses ubiquitous in nature and most of our adult population harbors multiple latent species (18). One of these is varicella zoster virus (VZV), an alphaherpesvirus responsible for chicken pox in children (varicella) or shingles (zoster) in elderly or immune compromised hosts (25). Though nucleoside analogs and a live-attenuated vaccine are currently available therapy against VZV, challenges include low bioavailability, toxicity and risk of viral resistance (31). Herpesviruses, along with other double stranded DNA viruses, replicate by packaging long concatamers of viral DNA through a “portal” protein into preformed procapsids in a process called encapsidation (11). The VZV portal protein is pORF54, an 87-kDa monomer that oligomerizes into a dodecameric ring (40). Alternate methods of combating infection could include blocking viral replication at the encapsidation step by inhibiting portal formation.

Mutations in VZV portal monomers were generated by inserting 5 amino acids randomly into ORF54, the gene coding for portal protein. A library of 55 different mutants with varying insertion sites were obtained and categorized by the suspected portal domain affected (clip, wing, crown, or tunnel-loop). pORF54 monomer mutants were evaluated through replication kinetics, exposure to thiourea compounds and intermolecular interactions between mutant monomers. The majority of the 55 mutant monomers were randomly generated in either wing or crown domains of portal monomers. Two mutants with amino acid insertions in the clip and stem domains (AA 305 and AA353 respectively) showed strong resistance to thiourea compound II suggesting the importance of that region for compound binding. Finally, monomeric mutants that failed to assemble into successful portal alone were able to replicate upon co-infection with different mutants. These data provide evidence that modifications to individual portal subunits can result in varying success of portal formation, drug resistance or susceptibility and ultimately viral replication.
INTERMOLECULAR COMPLEMENTATION BETWEEN pORF54 MUTANT MONOMERS AS A MECHANISM TO STUDY VARICELLA-ZOSTER PORTAL DOMAINS

by

ELLIE M. KORNFEIND

Approved:

Robert Visalli, Ph.D Date
Thesis Committee Chair

Richard McCann, Ph.D Date
Thesis Committee Member
Program Director

Jon Shuman, Ph.D Date
Thesis Committee Member

Jean Sumner, MD Date
Dean