# CALIFORNIA STATE SCIENCE FAIR
# 2017 PROJECT SUMMARY

**Name(s)**

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**Project Number**

**S0503**

**Project Title**

## Combating Embryonic Neurovirulence of Zika and Flaviviruses Using InSilico Phylogenetic Analysis and RNAi Gene Silencing

**Abstract**

**Objectives/Goals**

Recent outbreaks of Zika (ZIKV), Dengue (DENV), Chikungunya (CHIKV), and Ebola are neurovirulent. Trends in congenital and neurological manifestations (microcephaly, Guillain-Barré Syndrome, hemorrhagic fever) are worrying. The objective is 1)Investigate why recent ZIKV and flaviviruses are teratogenic and neurovirulent 2)Use RNAi Gene Silencing to identify siRNA molecules and target sites for inhibiting virus replication and neurovirulence.

**Methods/Materials**

The project was conducted in 6 stages: 1)Phylogenetic analysis in MEGA7.0 to study ZIKV mutations. 2)Identify degree of similarity (dS) among neurovirulent strains with 3'UTR RNA secondary structure analysis using RNAfold and Simtree. 3)Identify frequent patterns in neurovirulent flavivirus genomes with alignment free analysis in Python. 4)Pearson's Correlation and Spearman's Rho tests to correlate stage 3 pattern counts to degree of neurovirulence (dN) in other teratogenic viruses (Rubella, human cytomegalovirus, etc.) 5)Study viral cross-dependency by correlating ZIKV/microcephaly attack ratio (AR) to DENV/CHIKV AR. 6)Design siRNA molecules to inhibit viral replication/neurovirulence using siDirect. Perform in silico folding using mFold and calculate free energy of molecules.

**Results**

Neurovirulent ZIKV strains are derived from Asian clade, as shown by phylogenetic tree and ds(0.8406) between Brazil and Thailand strain RNA secondary structures. Mutations in prM, NS1, NS5 genomic regions increased occurrence of AGGTCA and other patterns in neurovirulent strains. AGGTCA Retinoic Acid Response Element (RARE) count correlated to embryonic neurovirulence in ZIKV ($p=0.000418$), other flaviviruses ($p=0.014$), and other neurovirulent viruses ($p=0.0179$). DENV AR correlated to ZIKV AR ($p=1.04E-9$) in humans but not to microcephaly ($p=0.85$). CHIKV AR did not correlate to ZIKV AR ($p=0.3955$) but correlated to microcephaly ($p=1.36E-6$). In NS5 siRNA molecules, GC% for neurovirulent strains was consistently ~42.86% and Delta(G) was ~-29.8.

**Conclusions/Discussion**

Results show that excess endogenous retinol may influence embryonic neurovirulence. Retinoic acid is a factor for regulating neural tube and Homeobox genes crucial for brain development. Correlation of RARE sequence count to dN indicate that mutations impacting RARE affect retinoic acid pathway and cause fetal malformations. siRNA molecules designed may help silence mutations and prevent embryonic neurovirulence.

**Summary Statement**

This project has identified mechanisms by which recent strains of ZIKV and other flaviviruses impair brain development and cause fetal malformations and provided potential siRNA molecules to silence viral replication and neurovirulence.

**Help Received**

My science teacher and research club mentor, Mrs. Segal provided valuable guidance. My parents provided encouragement.