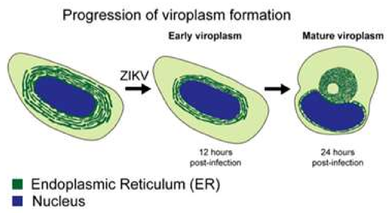
Zika virus NS3 drives the assembly of a viroplasm-like structure

Jenna Jones, Tania Sultana and Timothy Megraw

Background

Zika virus (ZIKV) is a flavivirus, a type of RNA virus. It originated from Uganda, Africa in the Zika forest. most known for its outbreak in the Americas during the 2016 Olympic games. While in some countries, cases of zika virus are almost nonexistent, however the virus still runs rampant in many areas of the world. Currently, there is no vaccine for the virus. ZIKV in patients particularly in pregnant patients has been shown to affect the brain development of the fetus.

Currently it is known that ZIKV encodes for 10 proteins:
7 structural and 3 nonstructural proteins. Additionally, it is known that some of the organelles are
affected by the formation of the viroplasm or replication organelle. The replication organelle is where the virus replicates its genome. Currently it is not known the specific proteins or mechanisms that are responsible for the formation of the replication organelle. This paper helps fill these gaps of knowledge by determining which protein is primarily responsible for the formation of the replication organelle. The results of this paper could help lead to the development of a vaccine for ZIKV.



Methods

This study focused on SNB19 cells, a type of human brain cell. We used RPMI media. Cells were transfected with FLAG-NS3 for 48 hours and fixed with methanol. The cells were infected with MR766, the African strain of ZIKV, to observe how the virus affects them. Some cells were also transfected with all ZIKV proteins individually to observe their role in viroplasm formation. To make the different parts of the cells easier to see, immunostaining was used. This staining method helped highlight both the virus and specific cell structures. High-resolution images were then taken using fluorescence microscopy to compare normal, infected, and transfected cells and to better understand how the virus changes the cells.

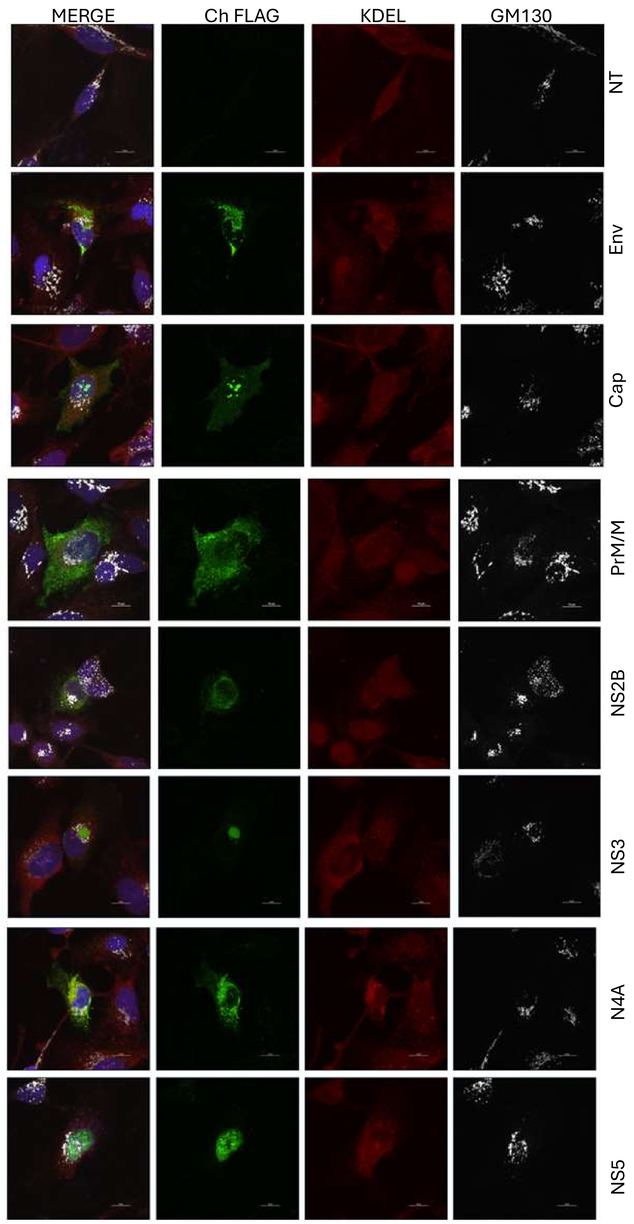


Figure: Nonstructural and Structural Proteins encoded by ZIKV

Results

NS3 is the main protein responsible for the formation of the replication organelle of Zika virus. NS3 was also found to recruit the ER and reorganizes the Golgi to the surface of the VLS. Among the structural proteins, EnV and Cap did not show Golgi or ER rearrangements. PrM/M showed some recruitment of the ER but no Golgi rearrangements. Among the nonstructural proteins, NS2B showed ER recruitment but no Golgi rearrangements. While, NS4A and 5 show neither.

Discussion

Some limitations are that while NS3 is a known main influence on the replication organelle formation, there may be other factors at play affecting it as well. Also, since ZIKV is a flavivirus, seeing how NS3 is related to the formation of the replication organelles in other flaviviruses as well is important. Future studies could look at both topics. In the future we are going to investigate NS3-driven VLS with ZIKV induced viroplasm to compare their features.

Acknowledgements

I want to thank the CRE program for the opportunity to participate in UROP. As well as my research mentor Tania Sultana for her unwavering guidance and support.

References

➢Buchwalter, R.A.; Ogden, S.C.; York, S.B.; Sun, L.; Zheng, C.; Hammack, C.; Cheng, Y.; Chen, J.V.; Cone, A.S.; Meckes, D.G., Jr.; et al. Coordination of Zika Virus Infection and Virus Infection and ViroplasmOrganization by Microtubukes and Microtubule-Organizing Centers. Cells 2021, 10, 3335. https:/doi.org/10.3390/cells10123335

➢White MK, Wollebo HS, David Beckham J, Tyler KL, Khalili K. 2016. Zika virus: An emergent neuropathological agent. Ann Neurol 80:479-89