

**TITLE:** EVALUATION OF THE ZIKA VIRUS EFFECT ON THE PRODUCTION OF ROS IN ASTROCYTE AND OLIGODENDROTIC PRECURSOR CELLS PRIMARY CULTURE.

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During the major Zika virus (ZIKV) epidemic in 2013 in French Polynesia and Brazil in 2015, neurological complications such as microcephaly and Guillain Barré syndrome were reported. According to the Brazilian Ministry of Health, until October 29, 2016, 10,039 cases of microcephaly were reported. It is known that one of the main causes of cellular damage in the Central Nervous System (CNS) is due to the increase and consequent accumulation of reactive oxygen species (ROS), which can be caused by viral infections, as described for Dengue, Japanese Encephalitis, Hepatitis C, among other viral diseases. Based on its susceptibility to ZIKV infection and its role in central nervous system homeostasis, astrocytes and oligodendrocytes precursor cells (OPC) provide a good cellular model for investigations of the mechanisms of entry of the virus and its effects. Being the primary culture of these cells, a tool used in this work in view of its greater similarity with the functions *in vivo* and at this stage, mouse brain developmental is equivalent to the early second trimester-stage of human CNS development. For the primary culture, neonate Swiss mice (1-4 days old) were used. The animals were euthanized, the brain removed and the cells enzymatically separated by addition of trypsin and mechanically with the aid of a serological pipette. After plating in cell culture bottles the glial cells were separated into astrocytes and OPC precursor cells by shaking. For quantification of ROS production, Sigma DCFDA reagent was used in microplate assay, the cells were then analyzed on a SpectraMax 4M fluorescence reader. The readings were taken at post-infection time intervals. The data show a gradual increase in ROS production over time, where there is a significant accumulation in the interval between 12 and 24 hours post infection. OPC show an even higher ROS production and accumulation compared to the other two analyzed cell lines (astrocytes and VERO). Analyzes made on BD FACS flow cytometer show that the number of cells with high production of ROS also increases from 3 hours of infection, where once again OPCs have a higher index than the other cells. These data suggest that cell types react differently to ZIKV infections and that a large accumulation of ROS occurs over the course of infection time.

**Keywords:** Zika Virus, Neuroglia Cells, ROS

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