Oncology 2025
The Cancer Control Challenge:
Better Understanding It to Better Face It

ANALYSIS OF mTOR PATHWAY EXPRESSION IN ORTHOFLAVIVIRUS ILHEUSENSE (ILHÉUS) INFECTION IN GLIOBLASTOMA

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Introduction: Glioblastoma (GBM) is the most aggressive primary brain tumor, characterized by high recurrence rates and limited therapeutic options. Oncolytic viruses (OVs) represent a promising therapeutic strategy due to their ability to selectively infect and destroy tumor cells. *Orthoflavivirus ilheusense* (ILHV), a neurotropic flavivirus, may exploit cellular pathways such as mTOR signaling to induce oncolytic effects. **Objectives:** To evaluate the oncolytic potential of ILHV in GBM by analyzing viral replication and the expression of genes associated with the mTOR signaling pathway. **Methods:** Vero E6 cells were used for ILHV stock preparation and viral titration ($TCID_{50}$). The GBM cell line AHOL1 was infected with ILHV at a multiplicity of infection (MOI) of 1 for 72 hours. Viral RNA was extracted using the Biospin Total RNA Extraction II Bioflux® kit, and RT-qPCR was performed with GoTaq® kits to quantify the expression of *mTORC1*, *mTORC2*, *Dicer*, *Drosha* and endogenous controls (*RPL1*, *RP38*). Statistical analyses were conducted using one-way ANOVA, Tukey's post hoc test and Pearson correlation analysis in Jamovi 3.2. **Results:** ILHV infection induced marked cytopathic effects in AHOL1 cells, peaking at 72 hours post-infection. Viral replication was significantly higher at 48 hours ($p < 0.05$). Expression levels of *Drosha* and *Dicer* decreased at 48 hours and increased at 72 hours, whereas *mTORC1* and *mTORC2* expression peaked at 72 hours (fold change > 2, $p < 0.01$). A negative correlation was observed between viral load, *mTORC2* and *Dicer* expression ($r = -0.65$, $p < 0.05$), while *Drosha*, *mTORC1* and *mTORC2* exhibited strong positive correlations ($r > 0.70$, $p < 0.01$). **Conclusion:** ILHV demonstrates significant oncolytic activity against GBM by modulating mTOR signaling and microRNA biogenesis pathways, resembling mechanisms described for other flaviviruses such as *Zika virus*, while potentially offering unique neurotropic advantages. As this is an in vitro study, translational applicability requires further validation. Future investigations using animal models are essential to confirm the efficacy and safety of ILHV as a therapeutic strategy for GBM.

Keywords: Oncolytic viruses; *Orthoflavivirus ilheusense*; mTOR signaling; microRNAs; Glioblastoma.