COMBATTING FUTURE BIOLOGICAL THREATS – HOST-DIRECTED INTERVENTIONS TO EMERGING THREATS FOR RAPID RESPONSE

Egr1 Influences Neuropathology In Mice Following Infection With Venezuelan Equine Encephalitis Virus VEEV) In A Sex-dependent Manner

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Emerging arboviruses, such as VEEV, are increasingly important causes of neurological disease in humans. After decades of research, there are still no FDA approved medical countermeasures available for VEEV and it is estimated that 4-14% of survivors of VEEV infection will develop chronic and debilitating neurological sequelae. Thus, there is an urgent need to identify potential therapeutic targets in order to develop therapeutics capable of limiting the neuronal insults encountered during and as a consequence of VEEV infection. Transcriptomic studies performed by our group revealed early growth response 1 (EGR1), an immediate early gene and transcription factor, to be significantly upregulated in human astrocytoma cells infected with VEEV. EGR1 is most notably known for its roles in neurological development and neuropsychiatric diseases but is also involved in regulation of inflammation, apoptosis, and antiviral signaling. Loss of EGR1 had no significant impact on viral replication, but resulted in decreased cell death and significantly altered the expression of numerous inflammatory response genes in vitro. Here, we evaluate the role of EGR1 in vivo using a mouse model of infection with VEEV TC-83. WT or EGR1 knockout (KO) C57BL/6J mice were intranasally infected with VEEV TC-83 and followed for survival, weight loss, and clinical symptoms of disease. Separate cohorts of mice were euthanized on 4, 7, and 28 days post-infection (DPI). Pathology was determined in the brain, serum samples were utilized for multiple cytokine/chemokine analysis, and bulk RNA sequencing was performed on brain samples. Loss of EGR1 had no significant impact on animal survival following VEEV infection, nor on viral RNA levels in the brain. Both WT and EGR1 KO male mice had significant pathology at 7 DPI including neuronal necrosis, meningitis, perivascular cuffing and neutrophil infiltration. At 7 DPI, female EGR1 KO mice had reduced neuropathology, specifically decreased meningitis. By 28 DPI, mice had clinically recovered from the infection; however significant neuropathology remained in all WT mice and in the male EGR1 KO mice. Importantly, little to no neuropathology was observed in female EGR1 KO mice at 28 DPI. Multiple proinflammatory cytokines/chemokines were altered in mice at 4 and/or 7 DPI infection. At 28 DPI, female EGR1 KO mice had decreased expression of IL-10, an anti-inflammatory cytokine, as compared to WT female mice. Differentially expression genes (DEGs) were analyzed using Ingenuity Pathway Analysis to identify altered canonical pathways. At 4 DPI multiple pathways involved in pathogen detection were altered whereas at 7 DPI there is a significant increase in T-cell and dendritic cell pathways, corresponding to the neuropathology observed. At 28 DPI, there is a shift to more adaptive immune response pathways being altered. Some notable DEGs between WT and EGR1 KO female mice at 28 DPI included Cox6a2, LRG1, and TMEM52. Collectively these results indicated that loss of EGR1 has limited impact on acute pathogenesis during VEEV infection, but impacts neuropathology resolution in female mice. Therefore, suppression of EGR1 is worthy of future investigation as a therapeutic target to reduce or prevent VEEV induced neuropathology and neurological sequelae in females.

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