REVOLUTIONIZING BIOMEDICAL RESEARCH: INTEGRATING CUTTING-EDGE AI/ML TO UNLEASH INNOVATION IN DRUG DISCOVERY AND THERAPEUTICS DEVELOPMENT

Comparative Analysis Of Equine Encephalitis Viruses (EEVS), Traumatic Brain Injuries (TBI), And Organophosphorus Nerve Agents (OPNA) As A Path To Neuroprotective Therapeutics

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Equine encephalitic alphaviruses (EEVs), traumatic brain injuries (TBI) and Organophosphorus nerve agents (OPNAs) are three diverse biological, physical, and chemical injuries which can lead to long-term neurological deficits in humans. Research on OPNA and EEVs is limited due to their classification as high-risk pathogens (BSL-3) and/or select agents. However, information can be gleaned by a comparative analysis with other intensively studied neuropathologies such as TBI. Disease manifestations of EEVs, TBIs, and OPNAs have notable commonalities including seizures, photophobia, paralysis, and coma, suggesting an overlap in pathway dysregulation. There are currently no publicly available antiviral treatments for EEVs, or neuroprotective drugs to combat the combined long-term neurological impact of TBI, OPNA, and EEVs which result in significant population health problems, increased health costs, and negative impacts on civilian and military personnel worldwide. We hypothesize that comparative neurological analysis of injuries from EEVs, OPNAs, and TBIs, can be utilized to identify potential medical countermeasures. To this end, we characterized the pathology, behavior, and transcriptomic changes caused by Venezuelan equine encephalitis virus (VEEV), sarin (OPNA), and moderate TBI in C57BL/6 mice. At both acute (Day 2 and 7), and chronic (Day 90) time points mice were sacrificed to look at pathology as well as immunohistochemistry analysis (IHC) for neuronal and microglia activation. We focused on the hippocampus due to it being a site of injury in all three neuropathological conditions and its importance for memory formation. Mice with TBI display interneuron loss over time in the dentate gyrus of the hippocampus. They also shown decreased Reelin expression, which is an extracellular protein that modulates synaptic activity in the brain. Likewise, we observed hilar interneuron loss, reduced expression of total Reelin, and reduced Reelin-expressing interneurons in mice at Day 7 post VEEV infection. Between the three different models, it has been established that emotional instability and behavioral changes are common post injury; therefore, we evaluated behaviors within mice utilizing well established behavior models. At day 30, 60, and 90 the behavior of the mice was assessed via Novel object Recognition and Y-Maze (memory) and Elevated Plus maze (anxiety). Both TBI inflicted and VEEV infected mice appear to have increased anxiety and memory loss at 60 and 90 DPI with fewer mice showing these deficits at 30 DPI. Our neuropathological analysis and behavioral testing has identified multiple similarities between VEEV infection and TBI. Finally, we performed single cell RNA-seq at acute and chronic timepoints to evaluate transcriptomic changes in the hippocampus, a site within the brain associated with memory and emotional regulation. Our data, along with published transcriptomic data were evaluated via a combination of pathway tools including gene ontology and Ingenuity Pathway Analysis. Transcriptomic analysis highlighted neuroinflammation, driven by proinflammatory markers IL -6 and TNF, as well as other potential drug targets such as S100 family member activation and Alpha-2-Macroglobulin (A2M) which may mediate neurological damage. Ultimately, we have identified several potentially druggable pathways to reduce long term neurological injury based on in house and published transcriptomic data.

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