more than 260,000 American have suffered mild traumatic brain injuries from sporting activities in the United States. Additionally, each year, as many as 3.8 million mild head injuries occur. It was determined that brain damage is occurring at all injury levels—varying degrees. This was done by observing the atrophy and damage after the administration of a traumatic brain injury of different severity.

Animals and Injury Model

For this research, mice were used as a study subject and the brain injury (mild, moderate, or severe) was administered using the 'Hit and Run' model.

Tissue Sampling and Preparation

Brain tissue samples were taken randomly at the 3, 7, 14, and 28 days after the administration. Samples were sliced and stained for imaging. DAPI stains for the nucleus, GFAP and CD68 expression were evaluated.

Imaging and Analysis

Images of the whole brain were taken with a fluorescence imaging microscope and were analyzed using Image J software. With this software, degrees of atrophy, and GFAP and CD68 expression were evaluated. Images of neurons and astrocytes were taken with a confocal microscope and analyzed for cell count using Image J software. This provided information on cell death for varying levels of injury at differing time intervals. The number of astrocytes was also counted to determine the correlation between cell death and glial activation.

Quantification of Atrophy:

Further analysis of the imaging data in the cerebral cortex and sub cortex indicates that no significant atrophy occurred with mild TBI (similar to a mild concussion), whereas the moderate and severe injuries show significant atrophy in both the cortices. Atrophy in the white matter, however, was significant at all degrees of injury severity at the 3 and 7 days marks. This significant amount of atrophy could be caused by brain sheering of the white matter due to the movement of the brain inside the skull during the administration of the injury, which helps strengthen this model for realistic data on TBIs.

Confocal Microscope Imaging:

The data collected from the confocal microscope imaging exposed a correlation between neuronal death and glial activation. With cell counting, it was determined that areas with higher concentrations of activated astrocytes had a lower number of neurons, further implying that the activation of astrocytes in even the mildest injury may correlate with neuronal death.

Results & Conclusion

Fluorescence Imaging:

Imaging analysis determined that GFAP and CD68 expression occurred in the cerebral cortex and sub cortex at all degrees of injury. The presence of GFAP and CD68 indicates the activation of glial cells, astrocytes and microglia in these areas, and thus, glial scarring occurred in even the mildest TBIs.

References


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