Characterization of cellular phenotypes through single-nucleus transcriptomic and epigenomic changes during TBI in young and aged mice

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Introduction: Each year, >27 million people experience a traumatic brain injury (TBI). TBI is associated with long-lasting adverse consequences, including an increased risk for neuropsychiatric and neurodegenerative diseases later in life. There is an urgent need to understand the mechanisms in the brain that contribute to these chronic effects of TBI. Here, we describe the cell type-specific transcriptional and epigenomic consequences of TBI in the hippocampus of mice in the controlled cortical impact (CCI) mouse model of TBI, using single-nucleus RNA sequencing (snRNA-seq) and single-nucleus ATAC sequencing (snATAC-seq).

Experimental design:

snRNA-seq; 90,872 cells from n=16 mice

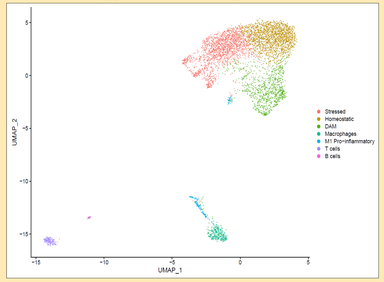
TBI		Sham	
18 mo	3 mo	18 mo	3 mo
4	4	4	4

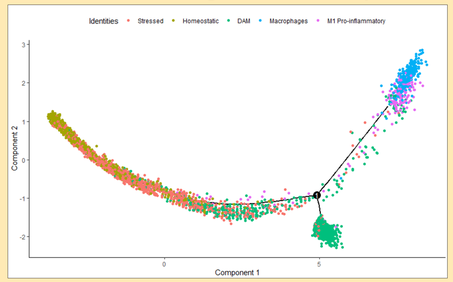
snATAC-seq; n=88,398 cells from n=8 mice

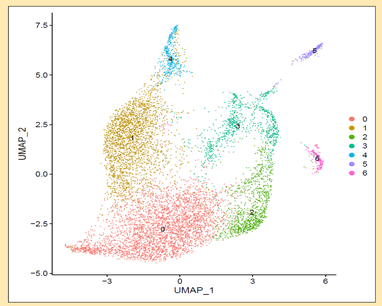
TBI		Sham	
18 mo	3 mo	18 mo	3 mo
2	2	2	2

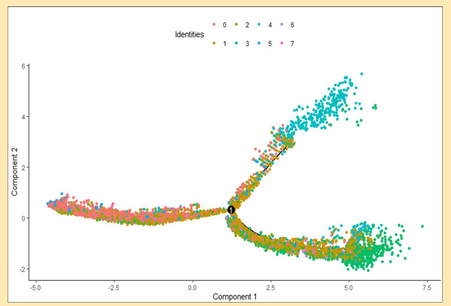
Background:

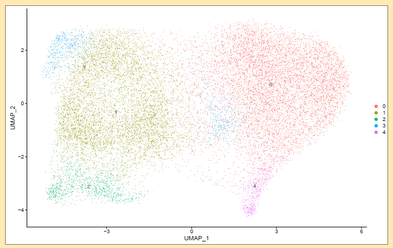
- Damage caused in the primary phase of TBI leads to chronic neurodegenerative processes leading to increased risk of psychiatric disorders like Alzheimer's disease (AD), depression, etc.[1]
- Cell-type specific changes occur as the neuroinflammatory effects mediated by microglia and astrocytes influence neuronal behavior.
- Newly characterized disease-associated microglia and astrocytes (DAM, DAA) in AD models are activated uniquely during neuroinflammation.[2,4]
- Exacerbated neurocognitive deficits have been observed in TBI affected aged mice compared to younger mice but difference in cell-type specific response has not yet been characterized.[4]
- Previous studies have also shown epigenetic predisposition of aged mice to disease but effects have not been characterized with respect to cell-type specific profiles.[5]

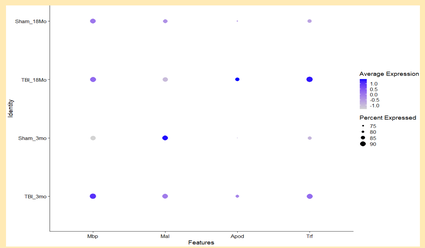












Endothelial cells

GO:BP	GO:BP
- Glial cell proliferation	- Synapse organization
- Gliogenesis	- Cell adhesion
- Localization	- Cell junction organization
- GO:CC - astrocyte end-foot, astrocyte projection	- Synapse assembly

Discussion:

- A total of 16 neuronal and non-neuronal clusters were identified from the snRNA-seq data; on integrating with snATAC-seq data, 11 clusters could be characterized.
- Microglia show two separate trajectories towards a neuroinflammatory state—DAM and M1.
- DAM cells seem to show enrichment for cell motility and localization genes as well as the TYROBP causal network activated during AD.
- Astrocytes also show two separate trajectories—corresponding to previously characterized DAA and a yet uncharacterized state which might be helping in neurogenesis.
- Oligodendrocytes show a more gradient-like change with strong effects of aging—mature oligodendrocytes seen majorly in 18 month old TBI mice compared to 3 month old counterparts.
- Endothelial cells show a signature correlating astrocyte recruitment for repair of blood-brain-barrier breakdown due to TBI.
- Next steps include integrating snRNA-seq and snATAC-seq data in a subtype specific way linking genes with accessible peak regions.
- Characterizing downstream effects of glial cell processes on neurons.
- Building gene regulatory network (GRN) to find out the regulatory features governing the changes in gene expression, which could lead to potential target.

References:

1.Saatman KE, Duhaime AC, et al. *J Neurotrauma*. 2008;25(7):719-738. doi:10.1089/neu.2008.0586
2.Keren-Shaul H, Spinrad A, et al. *Cell*. 2017;169(7):1276-1290.e17. doi:10.1016/j.cell.2017.05.018.
3.Hasel P, Rose IVL, et al. *Nat Neurosci*. 2021;24(10):1475-1487. doi:10.1038/s41593-021-00905-6.
4. Sun M, Brady RD, et al. *Brain Behav Immun*. 2019;80:536-550. doi:10.1016/j.bbi.2019.04.038.
5.Meng Q, Zhuang Y, et al. *EBioMedicine*. 2017;16:184. doi:10.1016/j.ebiom.2017.01.046.

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