BIOINF 545 Final Project Proposal

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1 Research Questions

Mast cells are myeloid immune cells found in connective tissue and mucosa that play an important role in innate immunity [4]. Over the course of development, mast cells are known to arise from two progenitor populations. Embryonic mast cells representing the early stage of mast cell development play a role in the early stages of immune development, and appear from primitive progenitor cells in the embryonic yolk sac [3]. Alternatively, definitive mast cells represent a more mature stage in mast cell development, arising from hematopoietic stem cells (HSCs) in the bone marrow and completing maturation in various tissues [3].

Hypothesis: Embryonic and definitive mast cells originating from distinct, lineage-specific progenitor populations rely on different transcriptional programs to determine mast cell fate during development.

1. What are the transcriptional programs that determine mast cell lineage fate in both embryonic and definitive development?
   (a) Our group will identify Differentially Expressed Genes (DEGs) related to transcriptional control/cell fate between embryonic and definitive mast cells.
   (b) We will use these DEGs to perform Gene Set Enrichment Analysis to determine if there are functional differences between definitive and embryonic mast cells.
2. What defines embryonic mast cell development from primitive myeloid hematopoietic stem cells?
   (a) We will perform pseudotime trajectory analysis using Monocole 3 to determine a differentiation trajectories for mast cell populations originating from primitive hematopoetic stem cells.

2 Dataset

2.1 Data type & number of observations/samples:

We will have two scRNA-seq datasets, one for embryonic and one for adult. We have narrowed down the original datasets to only the subcategory of mast cells for the purpose of this project. The embryonic dataset is derived from 10 samples from 4 to 8 postconception weeks (PCW) has 354 mast cells, and the adult dataset from 15 donors has 2238 mast cells.

2.2 Source of data:

- Embryonic:
  - Paper: Goh, I., *et al., Science.* 2023. [2]
  - Dataset
- Adult:
  - Paper: Tauber, M., *et al., Journal of Experimental Medicine. 2023* [1]
  - Dataset

3 Outline of Analysis Plan

Analysis Plan

1. Alignment and quality control for datasets in Cell Ranger.
2. Cell count matrixes will be processed and clustered with Seruat.
3. Thresholding will be applied to obtain DEGs from a pre-defined Mast Cell Cluster.
4. DEGs will then be input into Cytoscape for pathway analysis/Gene set enrichment.
5. Monocle for pseudo-time trajectory analysis on Cell Count Matrices Monocole 3.

4 Tools to be Used

- Cell Ranger
- Seruat
- Cytoscape
- Monocole 3

5 Allocation of Work

- Mariana: Background in stem cell biology and computational biology. Will help with writing background for the project and define mRNA markers for mast cells. Data analysis, R
- Chase: Scientific writing/editing, Data analysis and interpretation, R
- Christian: Background in Immunology and Mast Cell Biology. Data analysis and interpretation. Writing/editing.
- Ashley: Data analysis, R, scientific writing
- Paula: Data analysis, R, scientific writing

References

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[3] Gentek, R., Ghigo, C., Hoeffel, G., Bulle, M. J., Msallam, R., Gautier, G., Launay, P., Chen, J., Ginhoux, F., & Bajénoff, M. (2018). Hemogenic Endothelial Fate Mapping reveals dual developmental origin of Mast Cells. *Immunity*, *48*(6). https://doi.org/10.1016/j.immuni.2018.04.025

[4] St. John, A. L., Rathore, A. P., & Ginhoux, F. (2022). New Perspectives on the origins and heterogeneity of Mast Cells. *Nature Reviews Immunology*, *23*(1), 55–68. https://doi.org/10.1038/s41577-022-00731-2