Transcriptomic Analysis of Partial Reprogramming in the Aged Mammalian Neurogenic Niche

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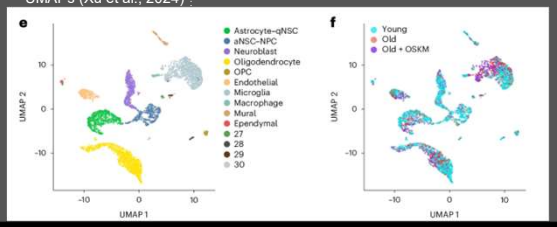
Introduction

- Subventricular Zone (SVZ) –Targeted and Whole-Body Partial Reprogramming was induced in mice to study effects of longevity by Xu, L. et al[1].
- Pulsed expression of transcription factors OCT4 (POU5F1 or OCT3/4), SOX2, KLF4, & c-MYC ('OSKM') were used to dedifferentiate cells in the SVZ.
- Single-cell RNAseq data from this experiment was analyzed to study the following:
  - **Transcriptional noise** of each cell-type within each age-treatment group
  - **Cell-type ratios** of each age-treatment group
  - **Transcriptional drift variance**[2] of each cell-type within each age-treatment group
- UMAPs (Xu et al., 2024)[1]:



Methods

- Three **Seurat Objects** for each mice cohort were downloaded into **Jupyter Notebook** from the source research paper[1] for R manipulation:
  - **SVZ-Targeted Cohort:** young control, old control, & old OSKM group
  - **Whole-Body Cohort 1**: old control & old OSKM
  - **Whole-Body Cohort 2:** young control, old control, & old OSKM group
- **NOTE: Whole Body Cohorts 1 and 2 were combined for analysis.**
- Used **gene count matrix** of each cohort to calculate **transcriptional noise** and **drift variance** for each.
  - Transcriptional Noise = sd(gene) / mean(gene)
  - Transcriptional Drift Variance (Rangaraju et al, 2015)[2]:

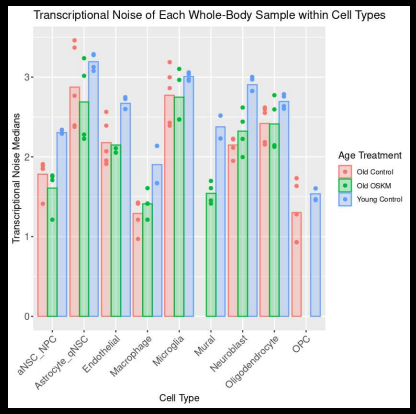
$$td_{gene\ x} = \left(\frac{\text{No.of transcripts}_{age[i]}}{\text{No.of transcripts}_{young\ reference}}\right) \qquad drift\ variance = \frac{1}{n-1}\sum_{i=1}^{n}\left(\text{td}_i - \overline{\text{td}}\right)^2$$

- Used **metadata** of each cohort to calculate **cell-type ratios** for each.

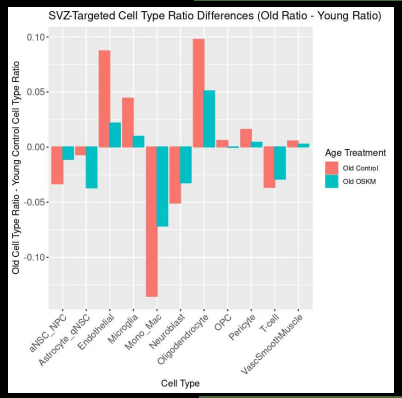
Hypotheses

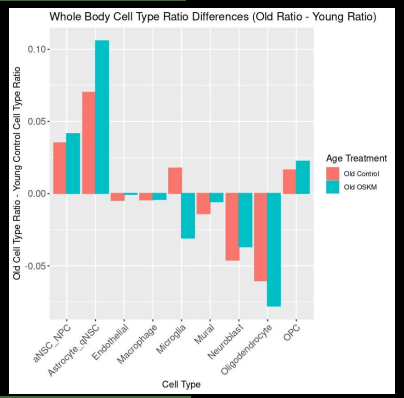
1. **Transcriptional Noise:** There will **not** be large differences in transcriptional noise with age. Nor will there be changes with OSKM.
2. **Transcriptional Drift Variance**: There **will be** large differences in transcriptional drift variance with age. This will be **reversed** with OSKM.
3. **Cell-Type Ratios:** There **will be** differences in cell-type balance with age (particularly less neuroblasts with age). This will be **reversed** with OSKM.
4. **SVZ-Targeted vs. Whole-Body**: The effects of OSKM will be the **same** between whole-body and SVZ-targeted.

Transcriptional Noise

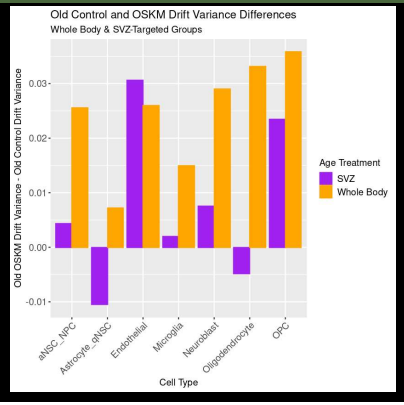


Cell-Type Ratio





Transcriptional Drift Variance



Results

1. **Transcriptional Noise:**
   - Young control samples consistently have **higher transcriptional noise** than the old control and old OSKM groups.
   - Old controls **have slightly higher transcriptional noise** than old OSKM groups in the SVZ-targeted group.
2. **Cell-Type Ratio:**
   - Old OSKM and young control groups consistently have **less cell-type ratio differences** than old control and young control groups in the SVZ-targeted cohort.
   - The **opposite effect** seems to take place in the whole-body group.
3. **Transcriptional Drift Variance:**
   - Old OSKM groups almost consistently **have greater drift variance** than old control groups in SVZ-targeted and whole-body cohorts.
   - SVZ-targeted old OSKM and old control drift variance difference **shows trend of being less than** whole-body old OSKM and old control drift variance difference.

Conclusion/Discussion

- Aging seems to have **decreased transcriptional noise** in this dataset and OSKM doesn't seem to alter this.
- SVZ-targeted partial reprogramming shows trends of **restoring cell-type balance to younger levels** than shown in old controls.
- Both SVZ-targeted and whole-body partial reprogramming shows trends of **increased drift variance across cell types.**
- Whole-body partial reprogramming is **not as effective as SVZ-targeted partial reprogramming** in restoring cell-type balance and transcriptional drift variance to younger levels.

Acknowledgements

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Sources

1. Xu, L., Ramirez-Matias, J., Hauptschein, M. *et al.* Restoration of neuronal progenitors by partial reprogramming in the aged neurogenic niche. *Nat Aging* **4**, 546–567 (2024). https://doi.org/10.1038/s43587-024-00594-3
2. Sunitha Rangaraju, Gregory M Solis, Ryan C Thompson, Rafael L Gomez-Amaro, Leo Kurian, Sandra E Encalada, Alexander B Niculescu III, Daniel R Salomon, Michael Petrascheck (2015) Suppression of transcriptional drift extends C. elegans lifespan by postponing the onset of mortality eLife 4:e08833 https://doi.org/10.7554/eLife.08833