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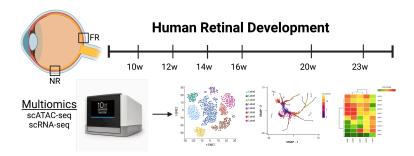


Figure 1A: The study design of this work. Samples were collected from either (1) whole retina at PCW 8, or (2) macula and periphery of the same donor's retina from PCW 10 to PCW 23. Subsequently, with a total of 28 samples from 14 donors, gene expression and open chromatin profiling from the same nuclei was performed using the 10X Chromium sn-dual-omic ATAC + Gene Expression technology (Supplementary Data 1). The bottom panel shows analysis workflow diagrams.



Step 1: Train **adult model** using the standard SCVI workflow.



Step 2: Load development data into the adult model.



Step 3: Train the **development model** so that adult data remain the same latent representation in both adult and model development model. Infer major class for development data.



Step 4: Remove adult data. Remove cells annotated as Microglia and RPE. Rerun UMAP with only development data.



Step 5: Run clustering and manually annotate retinal progenitor cells and Müller glia with established markers. Validate Annotation with inferred cell cycle.

Supplementary Figure 1A: Major class annotation workflow diagram. Step 1-3 are reference mapping with scvi-tools. In step 4 and 5, adult data was removed, and development data was clustered to annotate PRPC, NRPC, and MG based on established markers.



Step 1: Subset development data by major class.



Step 2: For each major class, merge development data with corresponding adult data with the same major class. Run UMAP and clustering for each major class.



Step 3: For each major class, manually annotate subclass and cell type. Leave the ambiguous clusters as precursors.

Supplementary Figure 2A: Subclass and cell type annotation workflow diagram. First, adult cells and development cells from the same corresponding major class were integrated together. Second, developmental cells were annotated manually based on gene expression similarities on UMAPs.



Step 1: Identify markers using adult data with differential gene expression analysis



Step 2: Check identified marker gene expression in development data to validate annotation by plotting dot plot.

Supplementary Figure 3A: Cell type annotation validation workflow diagram: First, marker genes were identified using adult data through a differential gene expression analysis for each major class. Subsequently, for each major class, developmental precursors and annotated data were visualized with those markers to illustrate gene expression patterns. Ideally, annotated development data should have similar gene expression patterns of those markers as adult data. The analysis was then tested using the method="t-test_overestim_var", which stands for overestimating the two-side variance in each group t-test. P-values were corrected using the 6Benjamini-Hochberg method, specifying corr_method="benjamini-hochberg".



Step 1: Compute RNA velocity for all cells and estimate a transition matrix based on RNA velocity



Step 2: Set mature neurons as terminal states, and for RPCs, compute fate probabilities towards terminal states.



Step 3: Run the Leiden clustering algorithm on NRPCs.



Step 4: Assign the cell fate by estimated fate probabilities for each NRPC cluster.

Figure 3A: Diagram of the NRPCs' fate probability inference workflow. Cell fate probabilities for each type of neuron were estimated based on velocity, and cell fate was assigned after performing cell clustering.

(B) UMAP of NRPCs colored by inferred fate. Cells for which cell fate cannot be determined are labeled in gray. The UMAP is specifically for NRPCs, distinguishing it from the global UMAP designed for all cells.



Step 1: Split cells by major class



Step 2: For each major class, fit two regression models with gene expression values.

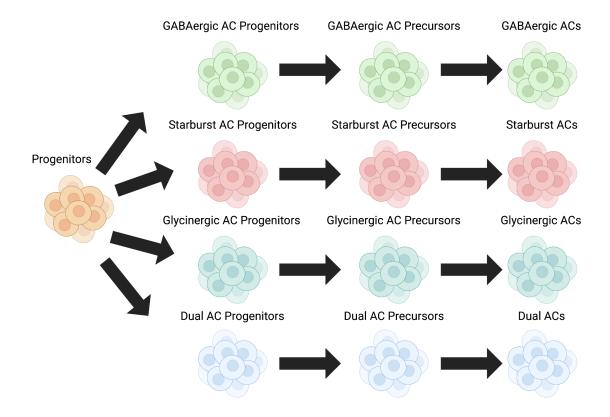
Model 1: Y~a*Location + b*Days Model 2: Y~c*Days



Step 3: Likelihood ratio test to compare Model 1 and Model 2

Supplementary Figure 9A: Differently expressed gene identification workflow diagram. For each major class, regression models were used for regression out the effects of "Days" during DEGs test between macula and peripheral. Then, a likelihood ratio test was performed for each group of cells by comparing the effects of "Region" and regressing out the effects of "Days", which is a vector of integer representing sample age (days post conception).





Supplementary Figure 7K: Hierarchical model for AC fate commitment during development of the human retina.

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Mythili Ravishankar, Customer Experience Associate
BioRender (Science Suite Inc.)

Mythili Ravishankar	23 June 2024
Signature	Date