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Supplemental information

Cochlear organoids reveal transcriptional programs of postnatal hair cell differentiation from supporting cells

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Supplemental Figures

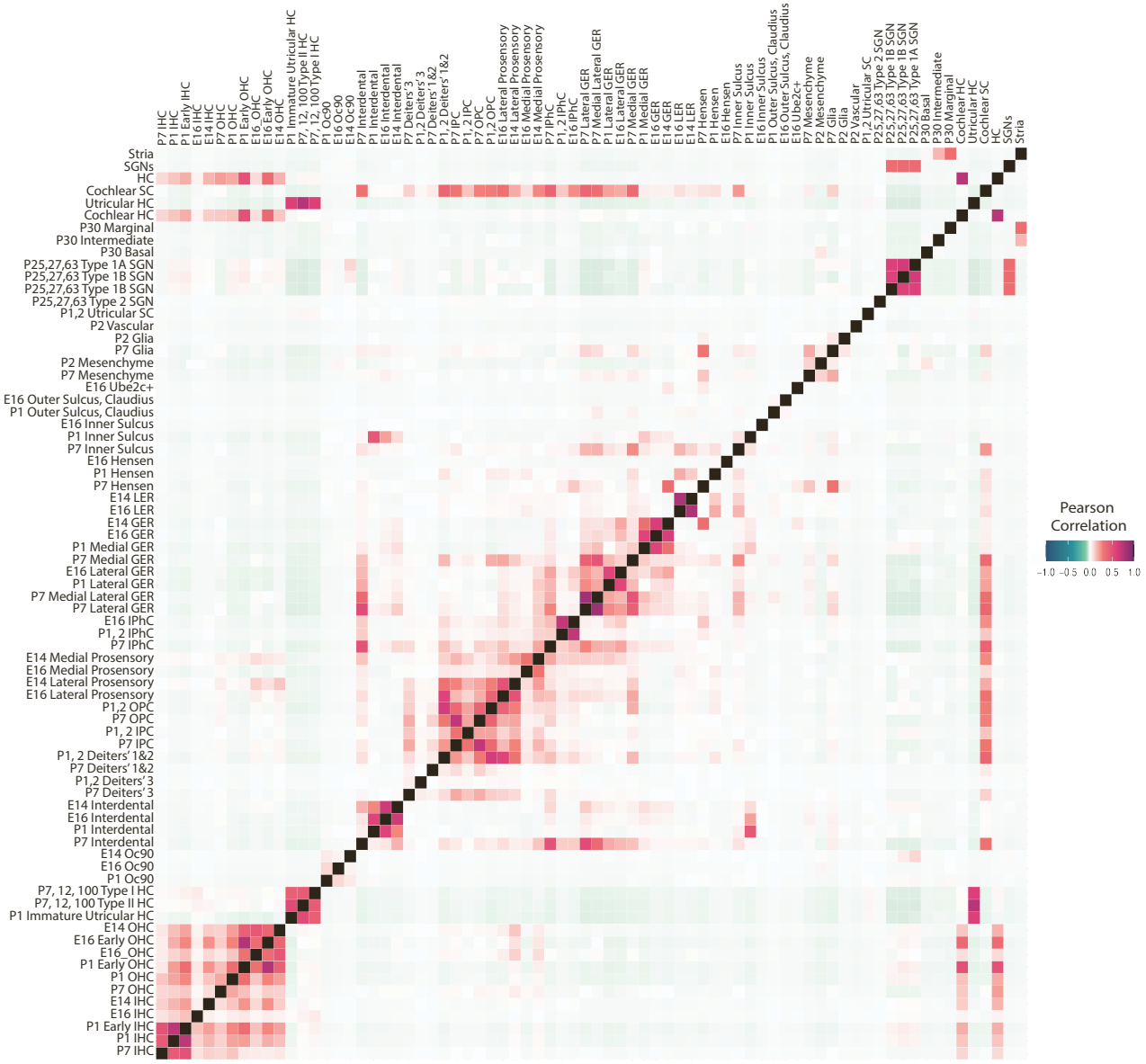

Figure S1 (related to Figure 1). Expression patterns of individual marker genes were obtained for each cell type. Gene expression patterns were used to calculate pairwise correlations of marker genes across cell types based on Pearson correlations of cell type specificity scores for up to 300 genes per cell type. Abbreviations used are: HC: Hair cell; SC: Supporting cell; IPhC: Inner phalangeal cell; IPC: Inner pillar cell; OPC: Outer pillar cell; GER: Greater epithelial ridge; LER: Lesser epithelial ridge; OS: Outer sulcus; SGN: Spiral ganglion neuron.

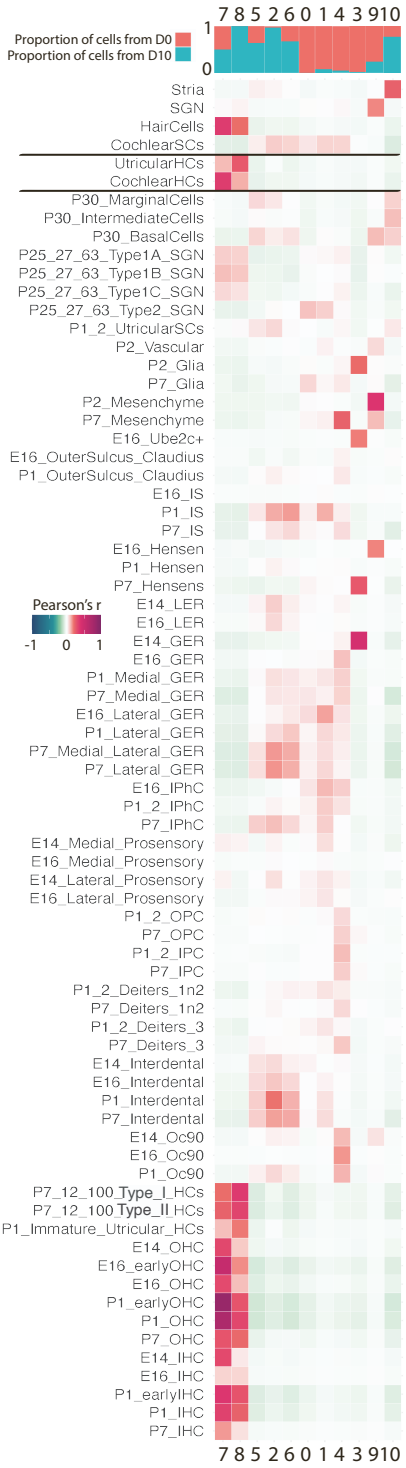

Figure S2 (related to Figure 2). The proportion of cells in each *in vitro* cluster was determined. Clusters from day 0 vs. 10 of differentiation are shown above the heatmap correlating expression of top marker genes from *in vivo* utricular and cochlear cell types to the genes expressed in each *in vitro* cluster.

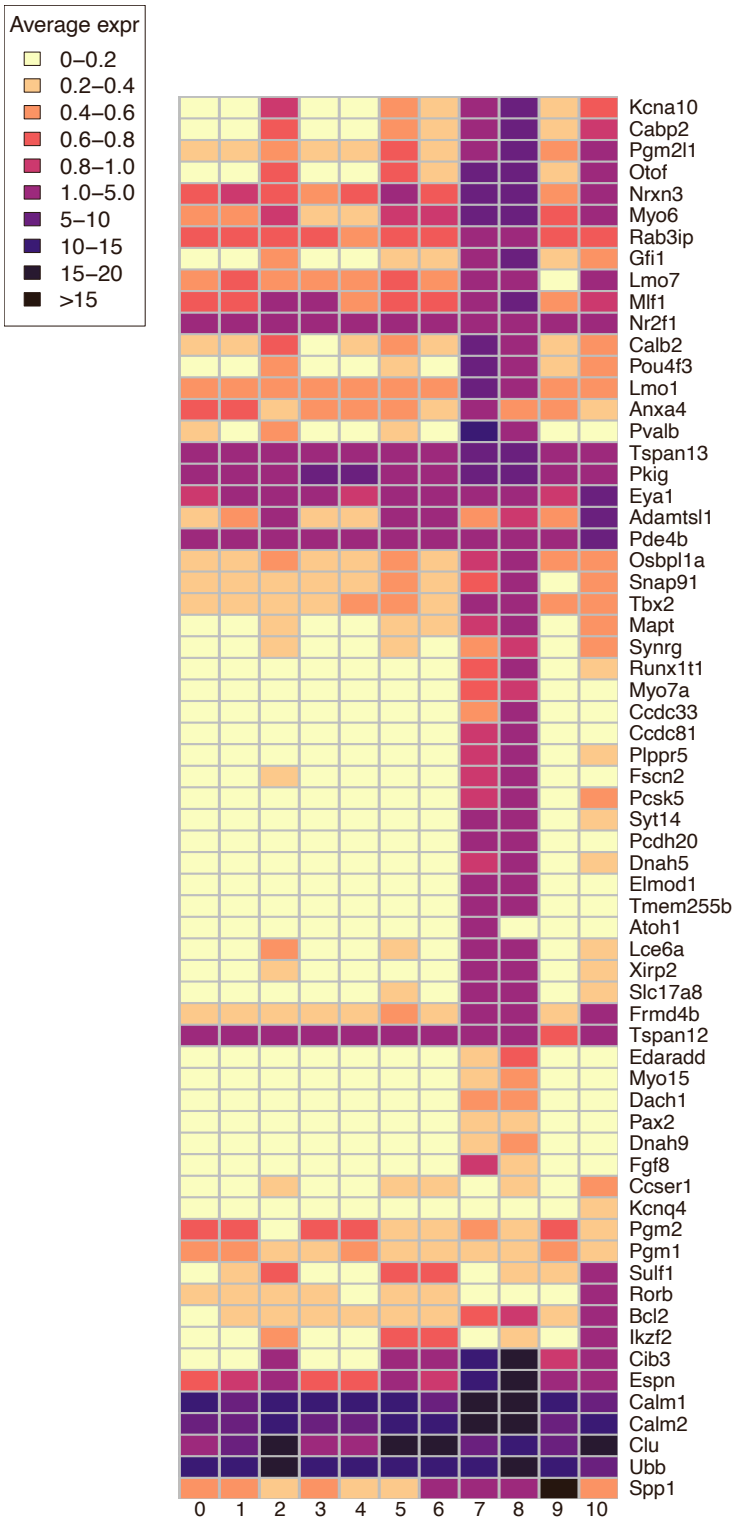

Figure S3 (related to Figures 2 and 3). A heatmap of hair cell-specific genes in all clusters of the single cell analysis was generated. Enriched expression of the hair cell genes was observed in clusters 7 and 8.

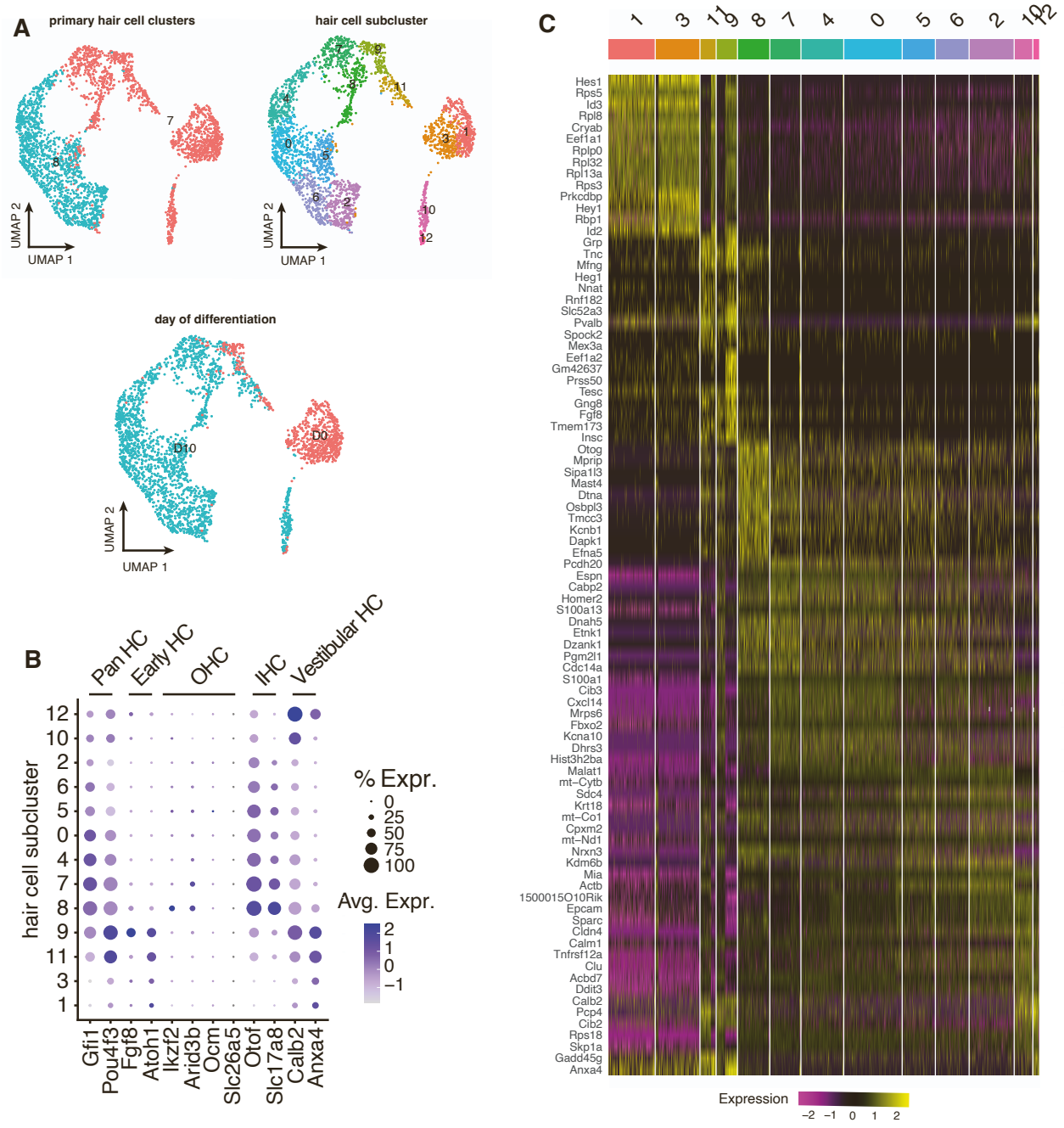

Figure S4 (related to Figure 2). Sub-clustering of the cells containing hair cell-specific genes (clusters 7 and 8) resolved thirteen clusters (Louvain resolution = 1.5). A. UMAPs labeled by sub-cluster identity, original cluster identity (7 vs. 8), and day of differentiation. B. Dotplot of markers for specific hair cell types across the thirteen clusters. C. Heatmap depicting the top marker genes for each of the 13 sub-clusters. HC = hair cell; IHC = inner hair cell; OHC = outer hair cell.

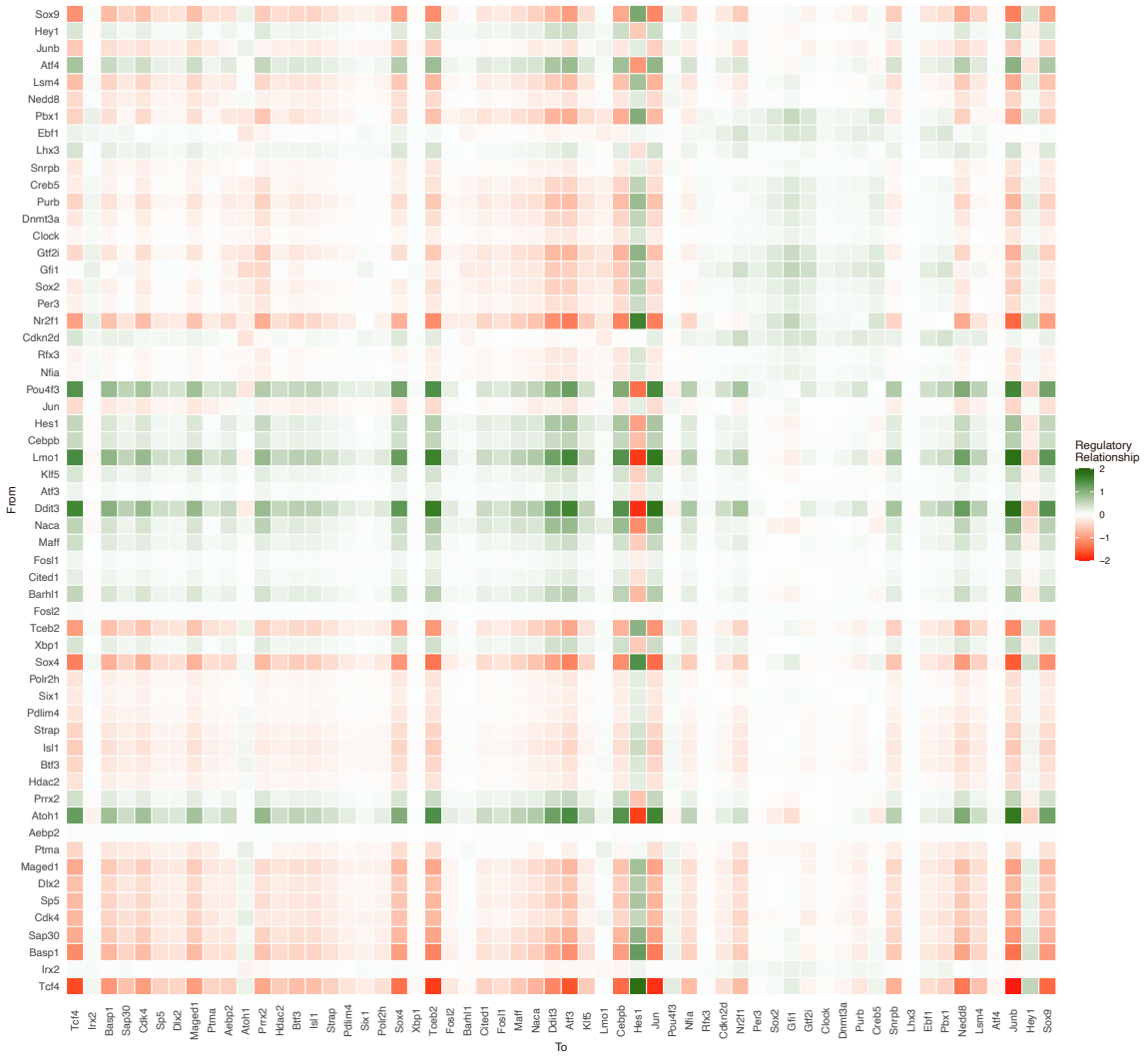

Figure S5 (related to Figure 5). A heatmap of the highly connected genes in the gene regulatory network is shown.

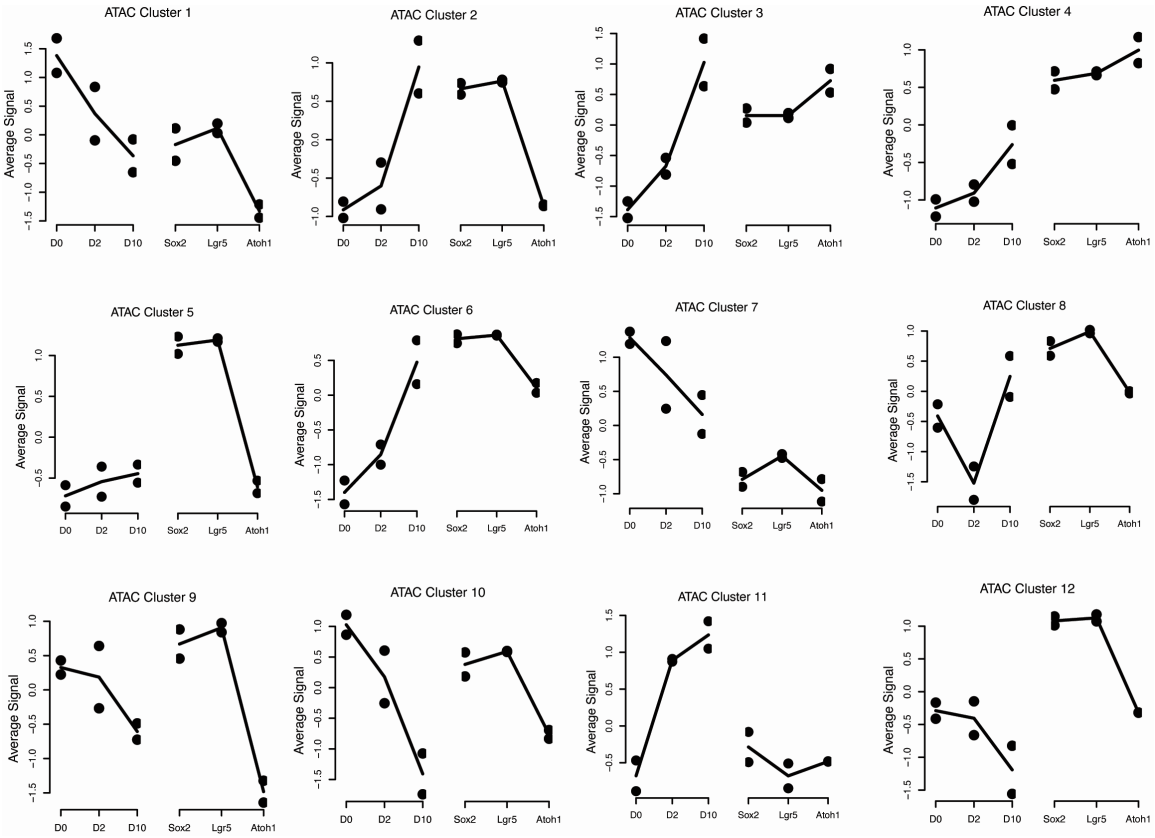

Figure S6 (related to Figure 6). Patterns of chromatin opening were ordered by K-means clustering. The data are from the ATAC-seq of the differentiating cochlear organoids at D0, D2 and D10.

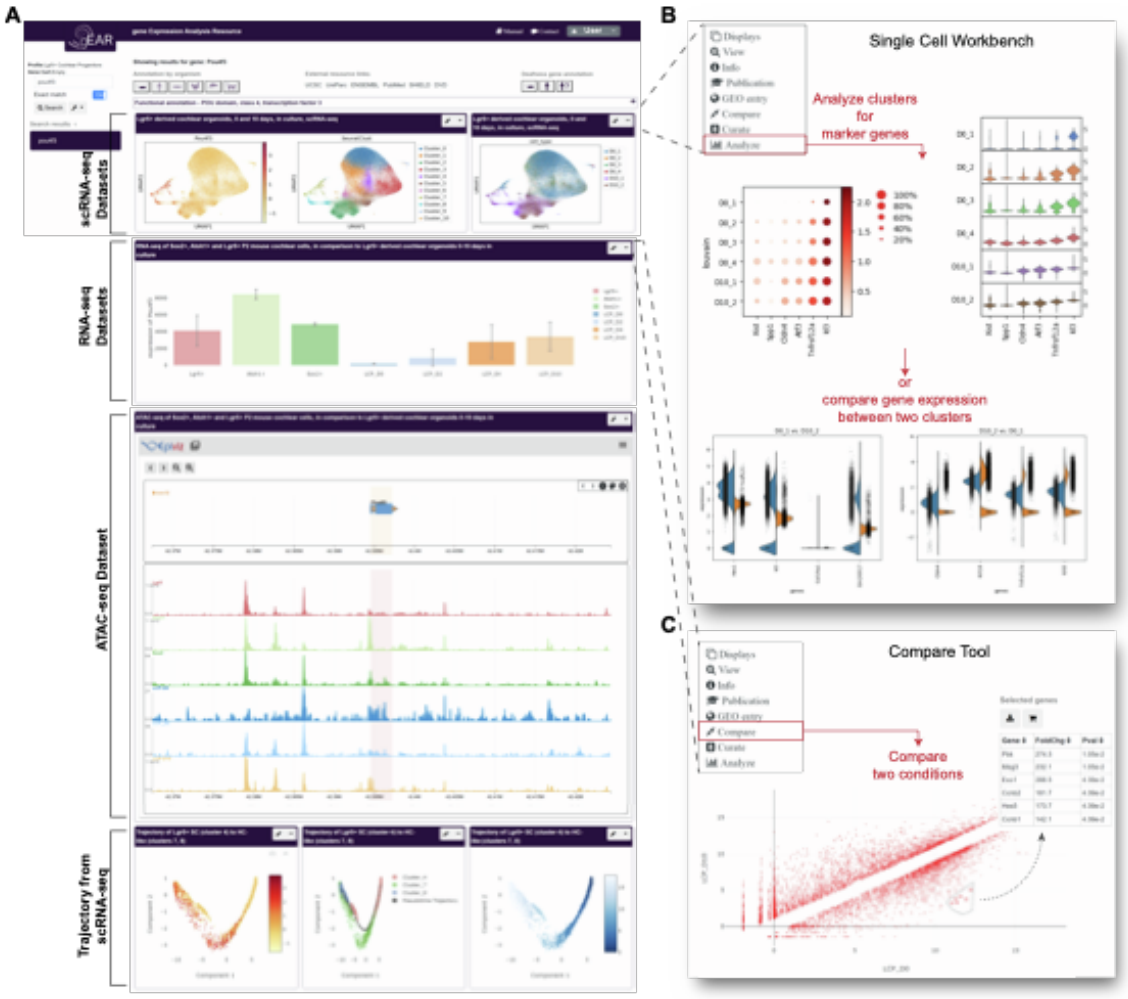

Figure S7 (related to Figures 2, 3 and 6). A custom profile was generated in the gEAR to support sharing, visualization and analysis of the processed data presented in this manuscript (https://umgear.org/Lgr5org). A. Overview of the manuscript profile, which contains the scRNA-seq data (UMAP), the RNA-seq data (bar graph), the ATAC-seq data (Epiviz) and trajectory. B. Example of the single cell workbench showing marker genes for the two time points across biological replicates (top) and the top 4 differentially expressed genes between day 0 replicate 1 (D0_1) and day 10 replicate 2 (D10_2) (bottom). C. Example of the use of the 'compare tool', showing the differentially expressed genes between D0 and D10 time points.