

Supplemental information

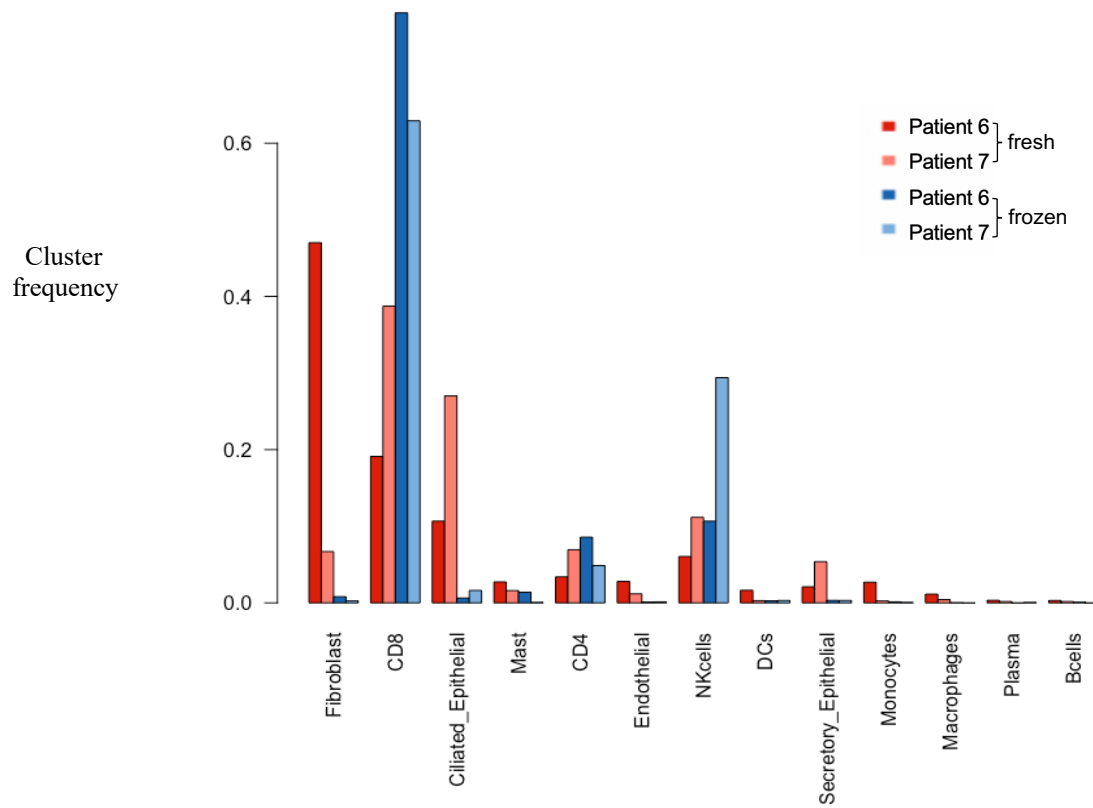
**Single-cell transcriptomics identifies gene
expression networks driving differentiation
and tumorigenesis in the human fallopian tube**

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Supplemental Figures

Dinh HQ*, Lin X*, Abbasi F*, *et al.*,

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B

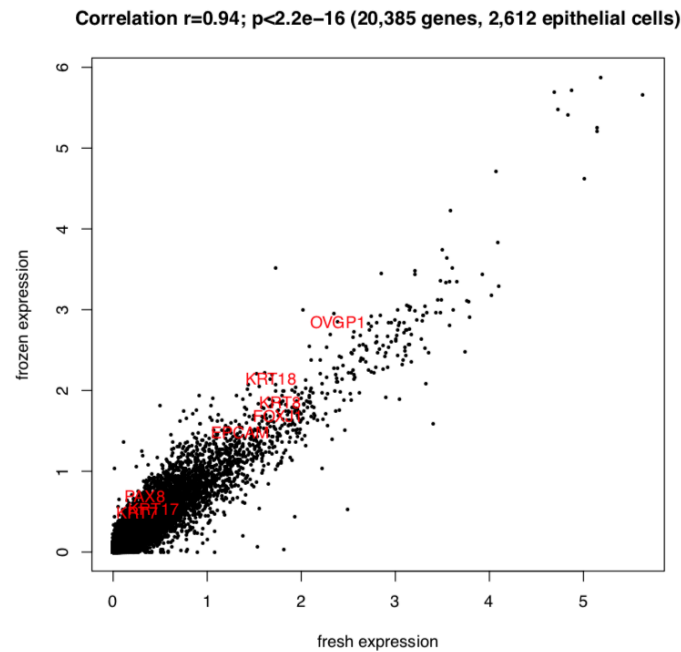


Figure S1. Related to Figure 1. The impact of cryopreservation on 10x single cell RNA-sequencing profiles of cellular composition and gene expression in the fallopian tube. (A) Major cell-type frequencies in two samples, determined from freshly processed tissues, and in the same samples profiled after cryopreservation. (B) Correlation of average gene expression in epithelial cells before ($n = 2,522$ cells) and after ($n = 90$ cells) cryopreservation.

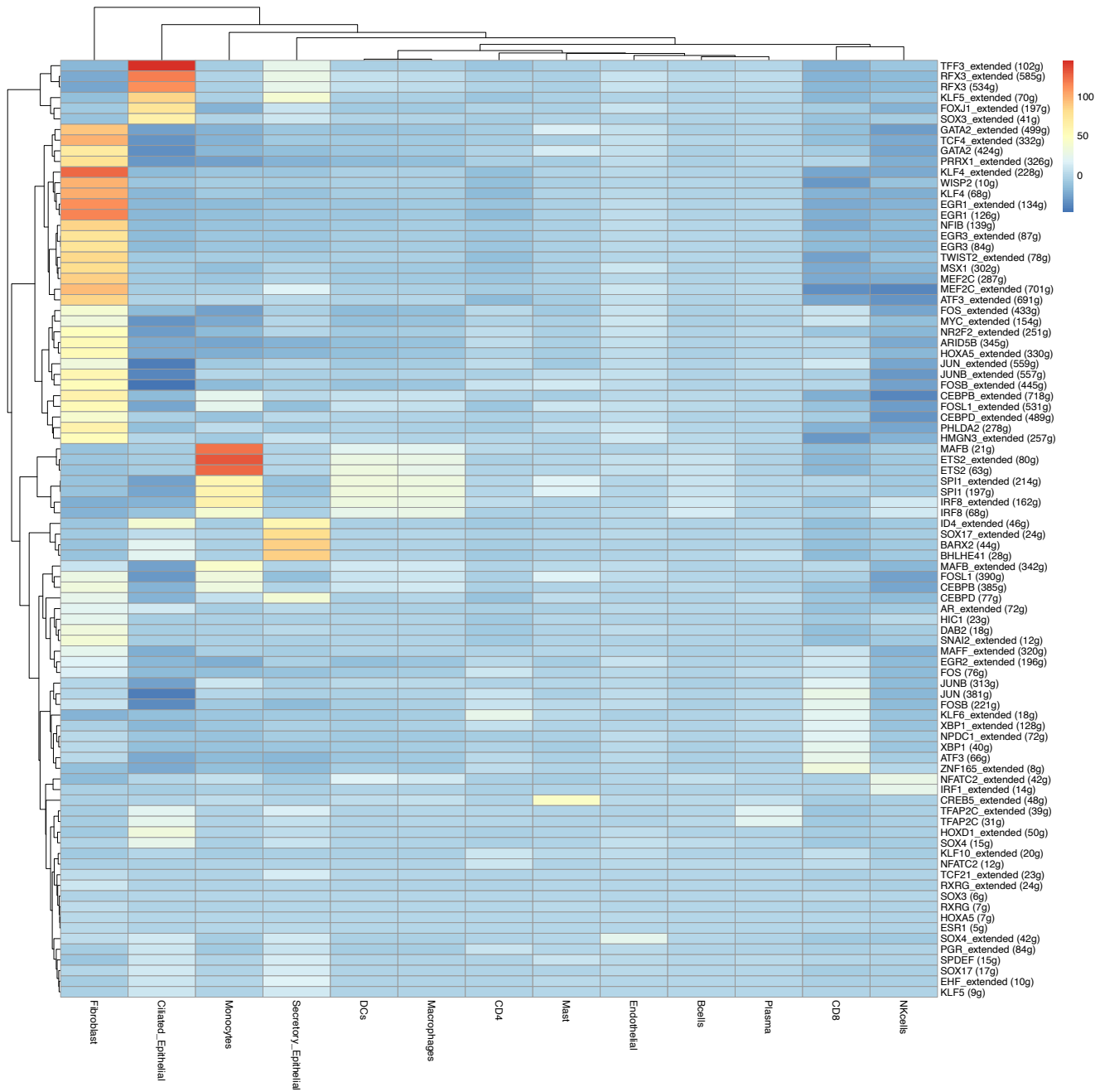


Figure S2. Related to Figure 2. Transcription factor regulons enriched in the major cell populations present in the human fallopian tube. TF regulons were identified by SCENIC analysis; cell-type-specific regulon enrichment is represented using the t-value from a linear model test of activity score between 1 cell-type *versus* the rest.

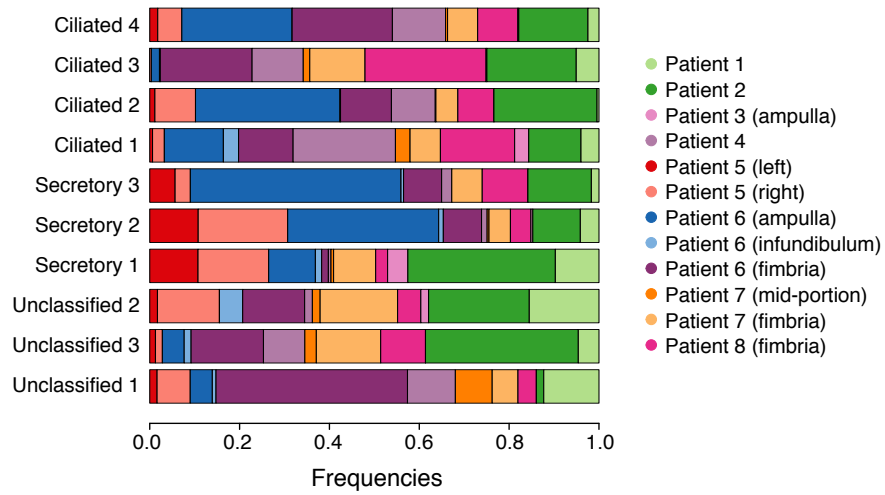


Figure S3. Related to Figure 3. Frequencies of epithelial sub-clusters by patient. Each cluster contains cells from all specimens in the cohort.

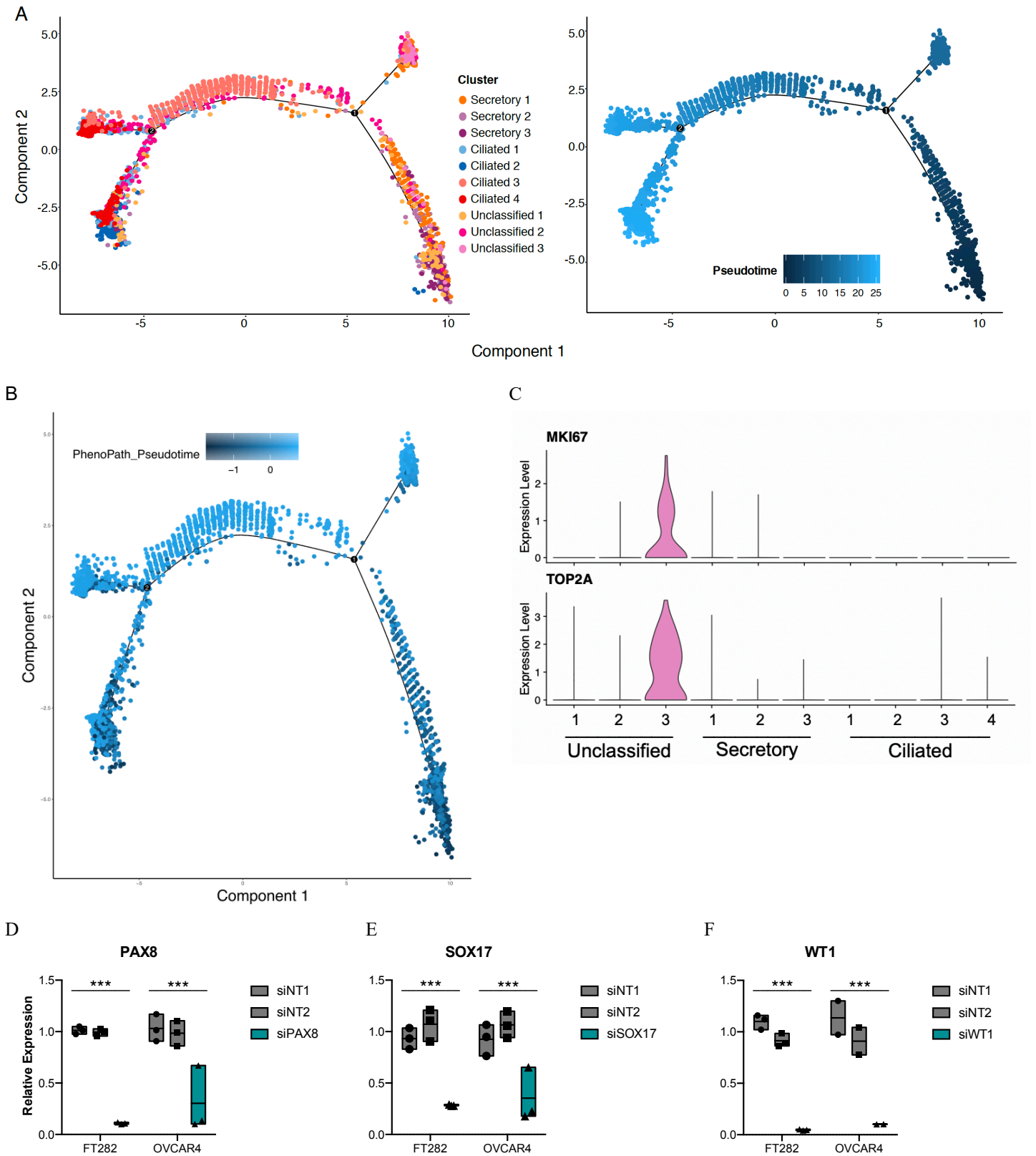


Figure S4. Related to Figure 4. Pseudotime analysis identified a proposed cellular trajectory for fallopian tube epithelial cells. (A) Alternative visualization for pseudotime analyses, performed using Discriminative Dimensionality Reduction with Trees (DDRTree) with the Monocle2 package. (B) Pseudotime analyses performed using PhenoPath closely correlate with time inferences obtained from Monocle2 (Pearson correlation coefficient $r = 0.35$, $p < 2.2 \times 10^{-16}$). Cells in unclassified cluster 3 are highly proliferative. Expression of *TOP2A* and *MIK67* (which encodes Ki67) are highest in unclassified cluster 3 compared to other epithelial clusters. High expression of *STMN1* and *MCM7* indicate that this population likely corresponds to cluster C9 from Hu *et al.* Validation of the relationship between secretory TFs and RUNX3 was performed using *in vitro* knockdown studies (Related to Figure 4H). (C)

Expression of representative proliferative markers (MKI67, TOP2A) is restricted to unclassified cluster 3 (D) *PAX8*, (E) *SOX17* and (F) *WT1* knockdown efficiencies in FT282, an immortalized fallopian tube secretory epithelial cell line, and OVCAR4, a prototypic high-grade serous ovarian cancer cell line. *** $p < 0.01$, two-tailed paired t-test. Data points on the floating bar plots show expression in three independent knockdown experiments performed at different passages (or two in the case of *WT1* knockdown in OVCAR4). Limits of the bars denote minimum and maximum relative expression; horizontal line indicates mean.

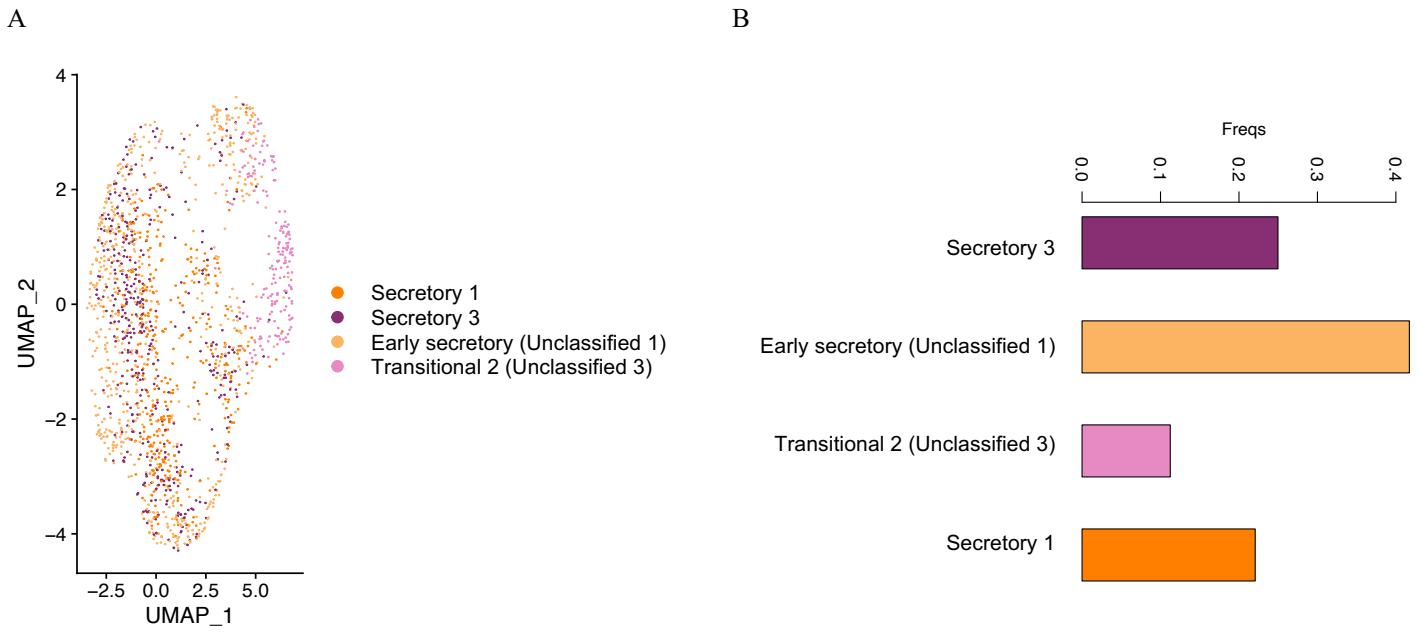


Figure S5. Related to Figure 5. Epithelial cell populations present in the immortalized FT282 cell line. The unclassified cluster 1 (renamed as ‘early secretory’) was the dominant cell type present in this model. (A) UMAP of 1,690 cells profiled, cells were labeled using 10 epithelial cell signatures derived from our human tissue data. (B) Histogram of frequency of epithelial cell types present in the FT282 cell line.

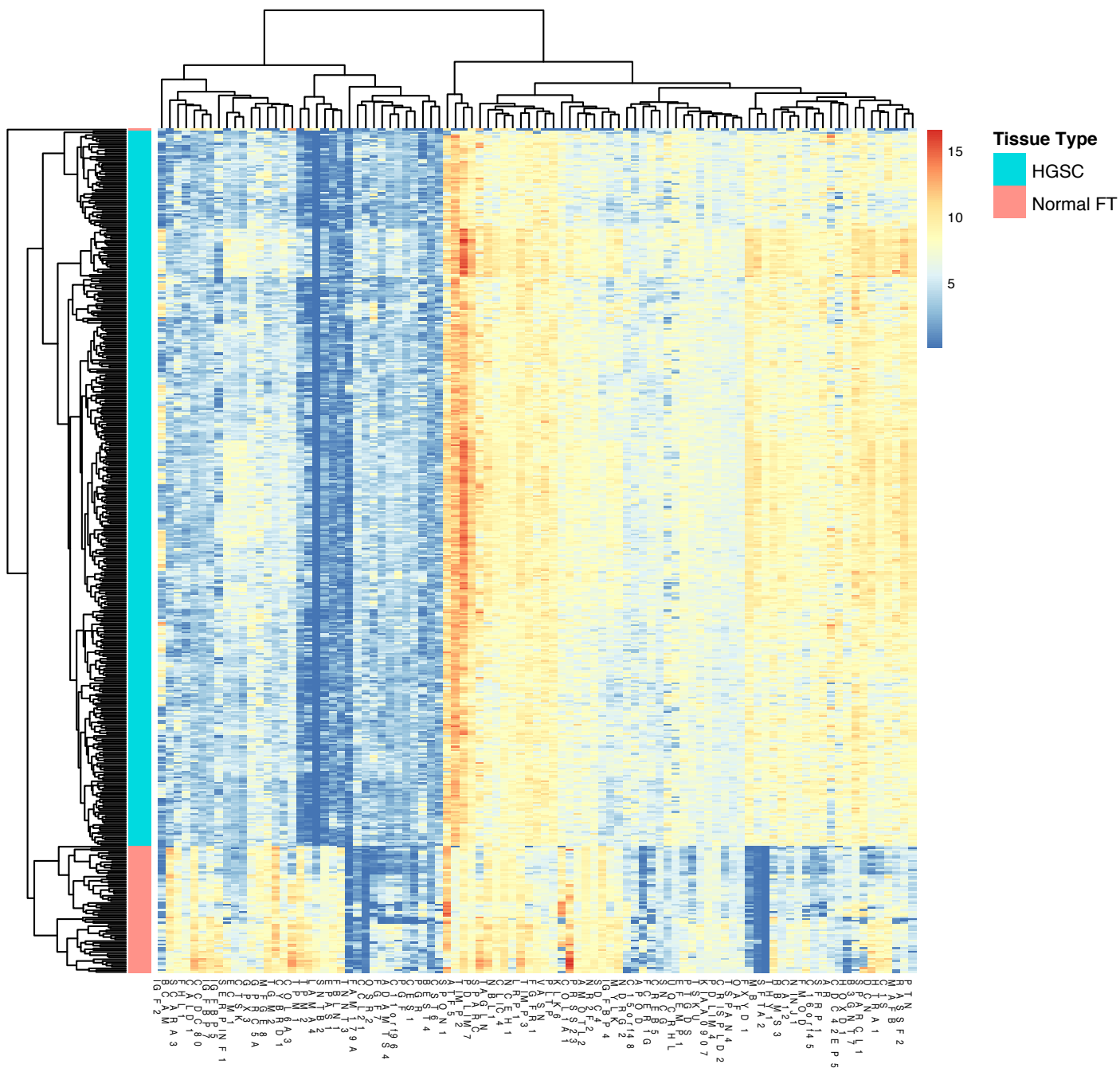


Figure S6. Related to Figure 5. Expression of genes in the NR2F2 regulon in 68 normal *ex vivo* fallopian tube cultures and 394 HGSCs. The NR2F2 regulon is enriched in early secretory cells, the major precursors for HGSCs, and is dysregulated during neoplastic transformation. The dendrogram was inferred using unsupervised hierarchical clustering and Euclidian distance.