Cell Reports, Volume *35*

Supplemental information

Contextual cues from cancer cells govern cancer-associated fibroblast heterogeneity

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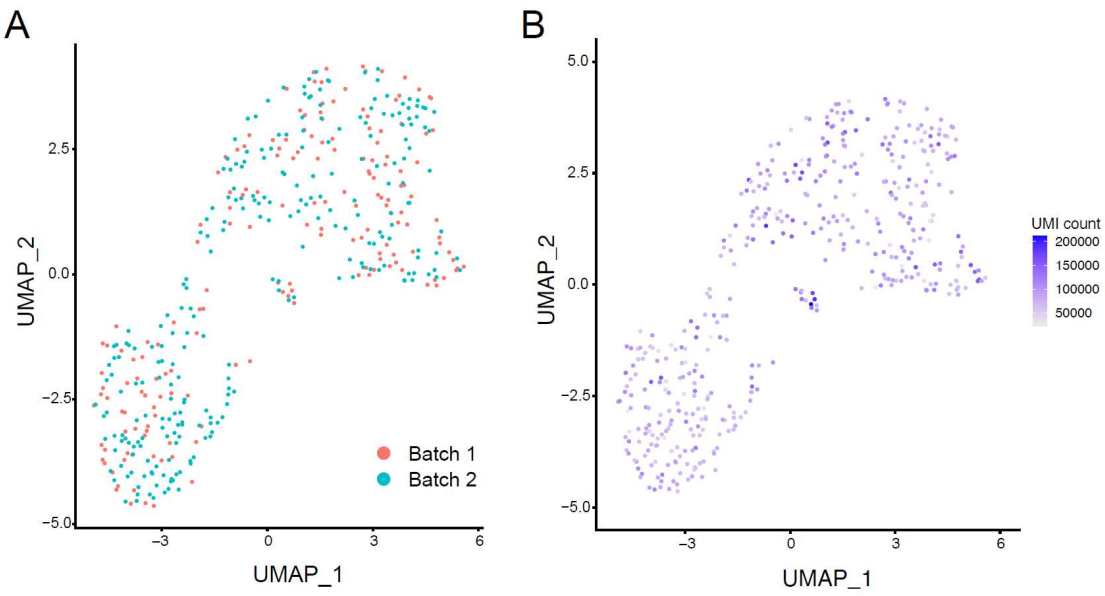
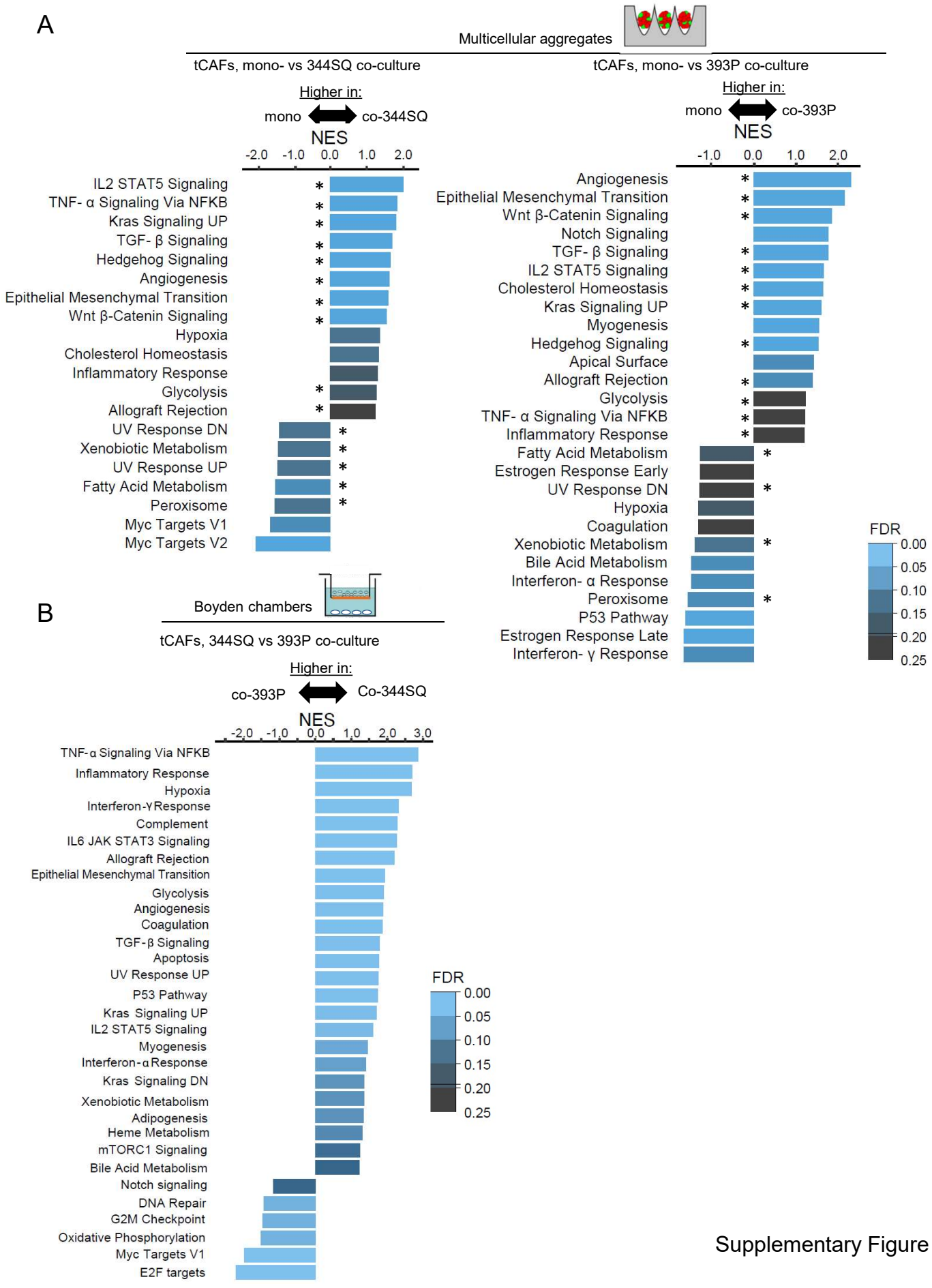


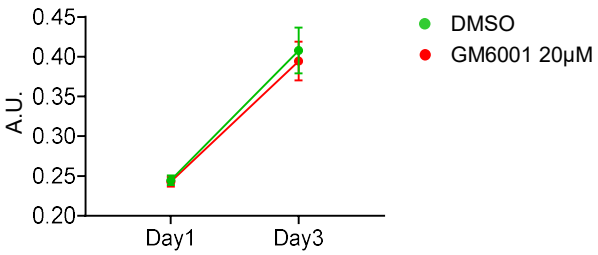
Supplementary Figure 1

Figure S1. Single-cell RNA sequencing quality controls. Related to Figure 1. (A, B) PCA plots depict batch #1 and #2 (A) and Unique Marker Identifier (UMI) numbers in each cell (B).



Supplementary Figure 2

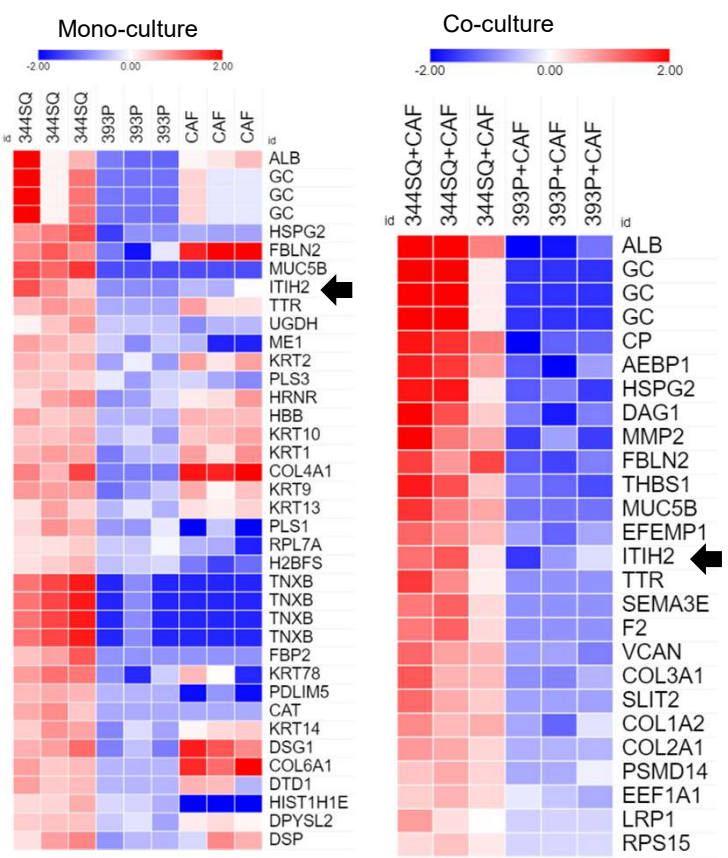
Figure S2. LUAD cells at either end of the EMT spectrum shape tCAF heterogeneity. Related to Figure 2. (A) GSEA of upregulated genes in tCAFs that were mono-cultured (mono-) or co-cultured with 344SQ cells (co-344SQ) or 393P cells (co-393P) in multicellular aggregates. n= 3 biological replicates per condition. (B) GSEA of upregulated genes in tCAFs that were co-cultured with 344SQ cells (co-344SQ) or 393P cells (co-393P) in Boyden Chambers. NES represented by bar length. FDR values color-coded. n= 5 biological replicates per condition. Hallmarks that were shared between Boyden chambers and multicellular aggregates are indicated (A, asterisks).



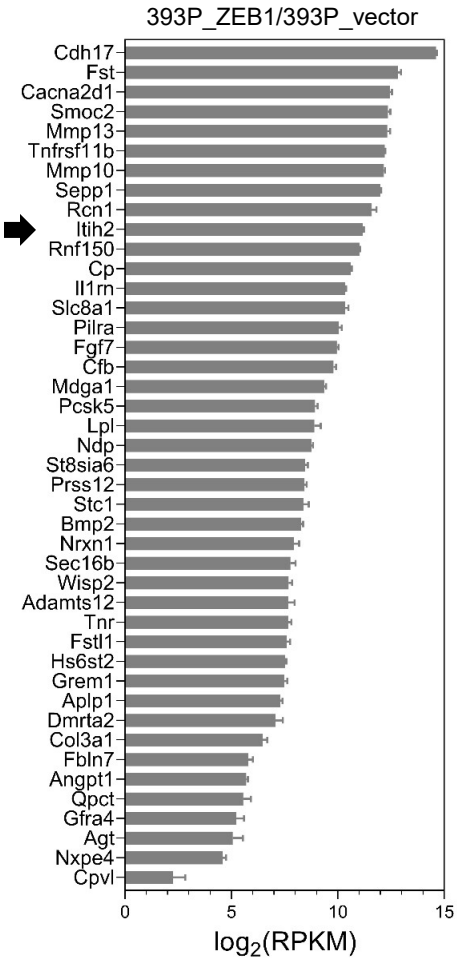
Supplementary Figure 3

Figure S3. Relative densities of tCAFs treated for indicated time periods with 20μM GM6001 (red) or vehicle dimethylsulfoxide (blue) determined by WST-1 assay. Related to Figure 4. A.U.: Absorbance units. n=3 biological replicates per condition.

A



B



Supplementary Figure 4

Figure S4. Identification of ZEB1-driven secreted factors. Related to Figure 6. (A) Heat map depiction of selected secreted factors identified by liquid chromatography-mass spectrometry analysis of conditioned medium samples from multicellular aggregates containing mono-cultured cells (left heat map) or co-cultured cells (right heat map). All proteins depicted had a P value < 0.05 (t-test). n=3 biological replicates per condition. (B) Relative levels of selected secreted factors identified by bulk-cell RNA sequencing (Tan et al., 2018). Values expressed as a ratio (393P_ZEB1/393P_vector). Log2FC: Log2 of fold change. n=3 biological replicates per condition