

Supplementary Materials

Supplementary Figure 1. Quality control metrics and batch effect correction in single-cell RNA sequencing data.

Supplementary Figure 2. Weighted gene co-expression network analysis reveals functional modules and their prognostic associations.

Supplementary Figure 3. Cell-cell communication landscape in the gastric cancer microenvironment.

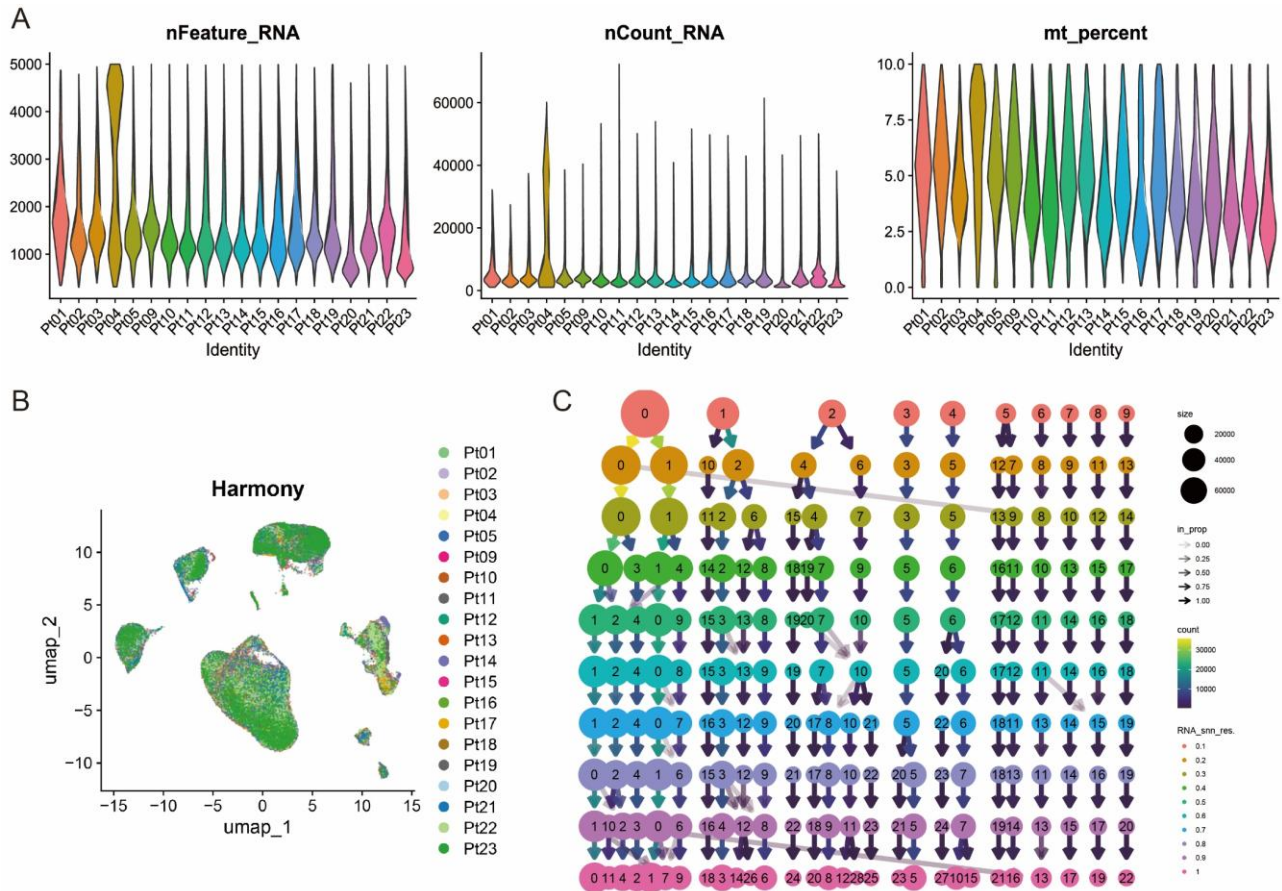
Supplementary Figure 4. Cell type distribution and functional gene clustering analysis.

Supplementary Figure 5. Differential cell-cell communication patterns across tissue types.

Supplementary Figure 6. CytoTRACE analysis and gene expression dynamics across tissue types and B cell phenotypes.

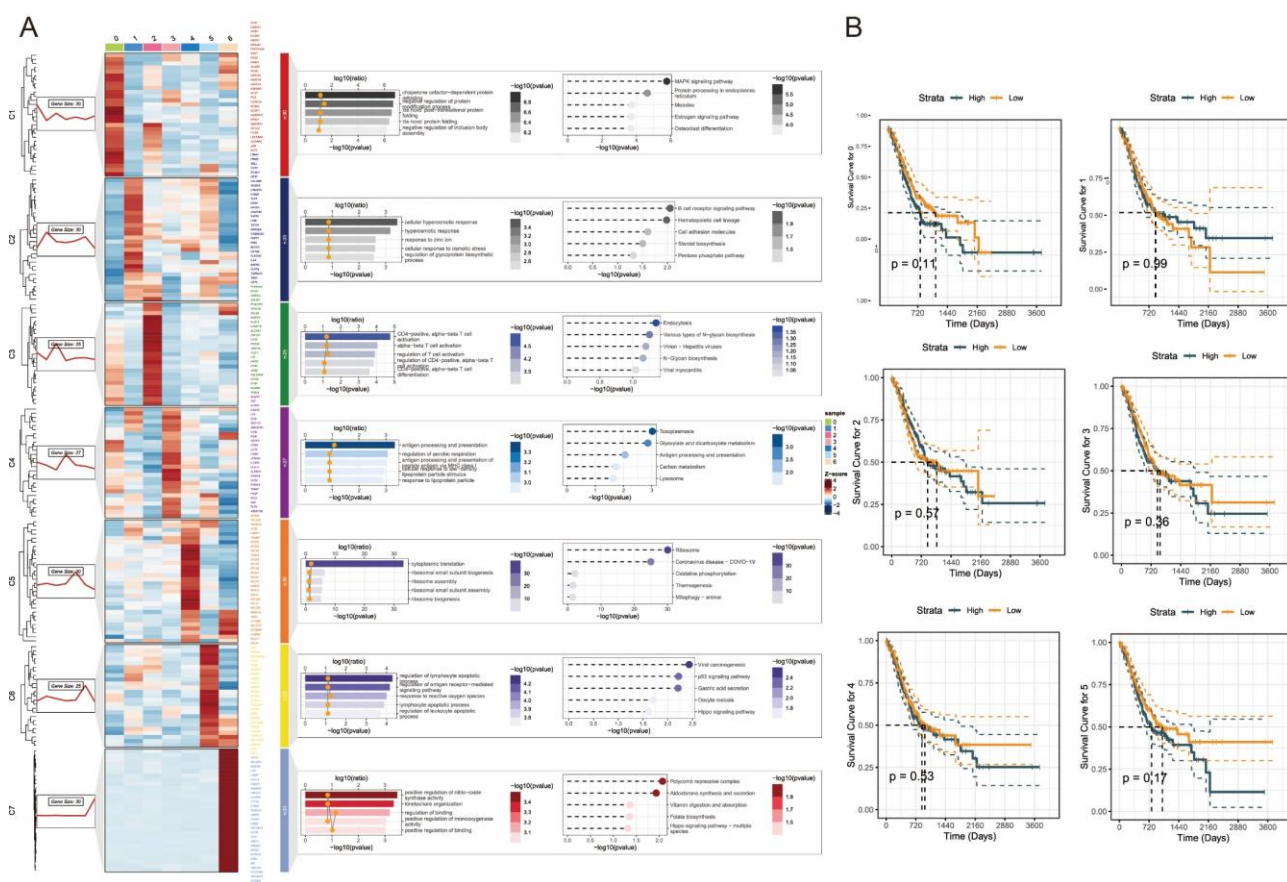
Supplementary Figure 7. Pathway enrichment and ligand-receptor interaction analysis in metastatic sites.

Supplementary Figure 8. KEGG pathway enrichment analysis of the turquoise module genes.



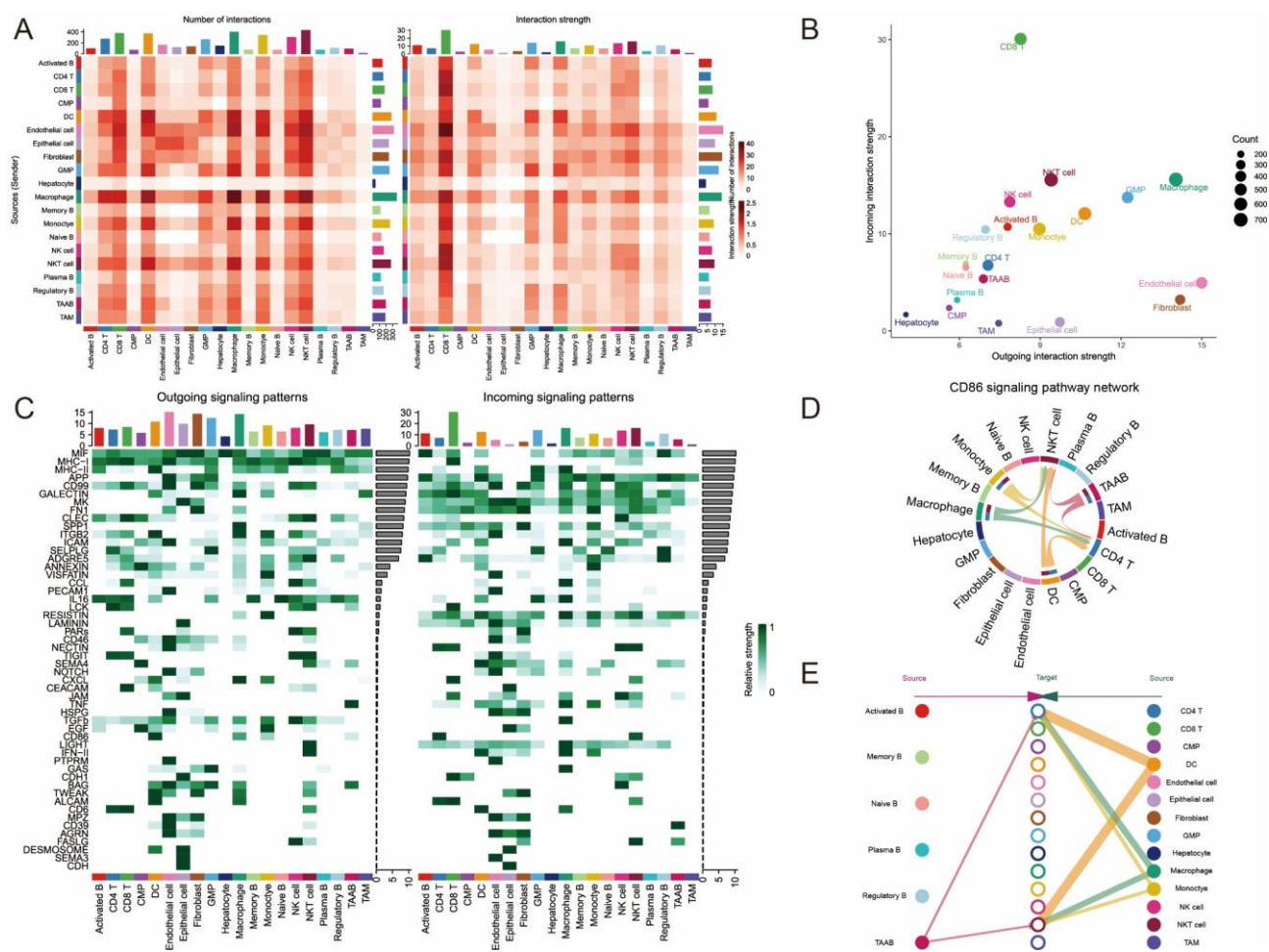
Supplementary Figure 1. Quality control metrics and batch effect correction in single-cell RNA sequencing data.

(A) Violin plots showing quality control metrics across patient samples, including nFeature_RNA (detected genes per cell), nCount_RNA (total UMI counts), and mt_percent (mitochondrial gene percentage). (B) UMAP visualization after Harmony batch correction demonstrates effective integration of cells from 20 patient samples (Pt01-Pt23). (C) Alluvial plot illustrating clustering stability across different resolution parameters, with node size representing cell count and arrows indicating cluster relationships.



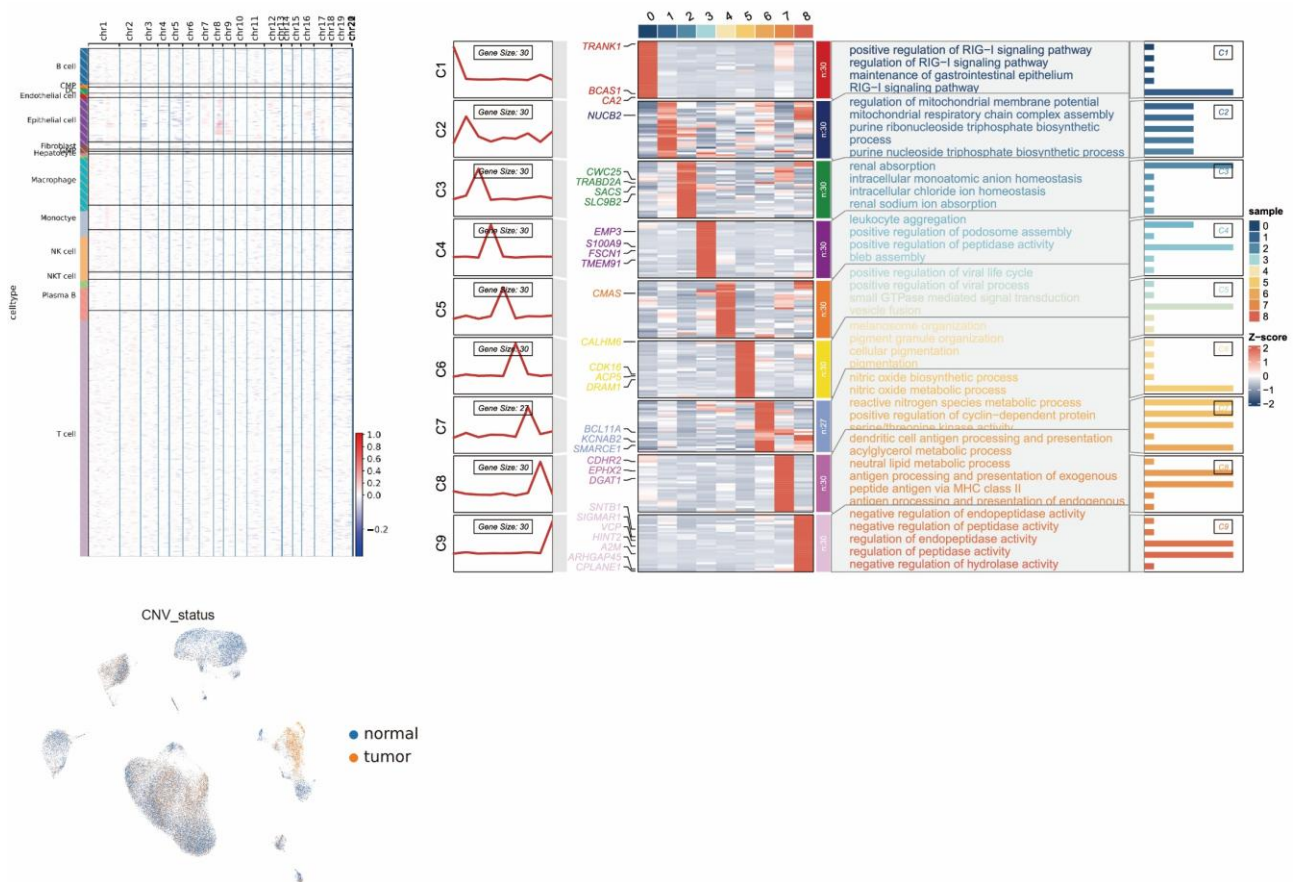
Supplementary Figure 2. Weighted gene co-expression network analysis reveals functional modules and their prognostic associations.

(A) Heatmap displaying gene expression patterns with hierarchical clustering identifying distinct co-expression modules (color bars on the left). Each module is functionally characterized through GO and KEGG enrichment analysis. **(B)** Kaplan-Meier survival curves for representative gene modules, stratifying patients into high and low expression groups. Most modules show no significant prognostic associations ($p > 0.05$).



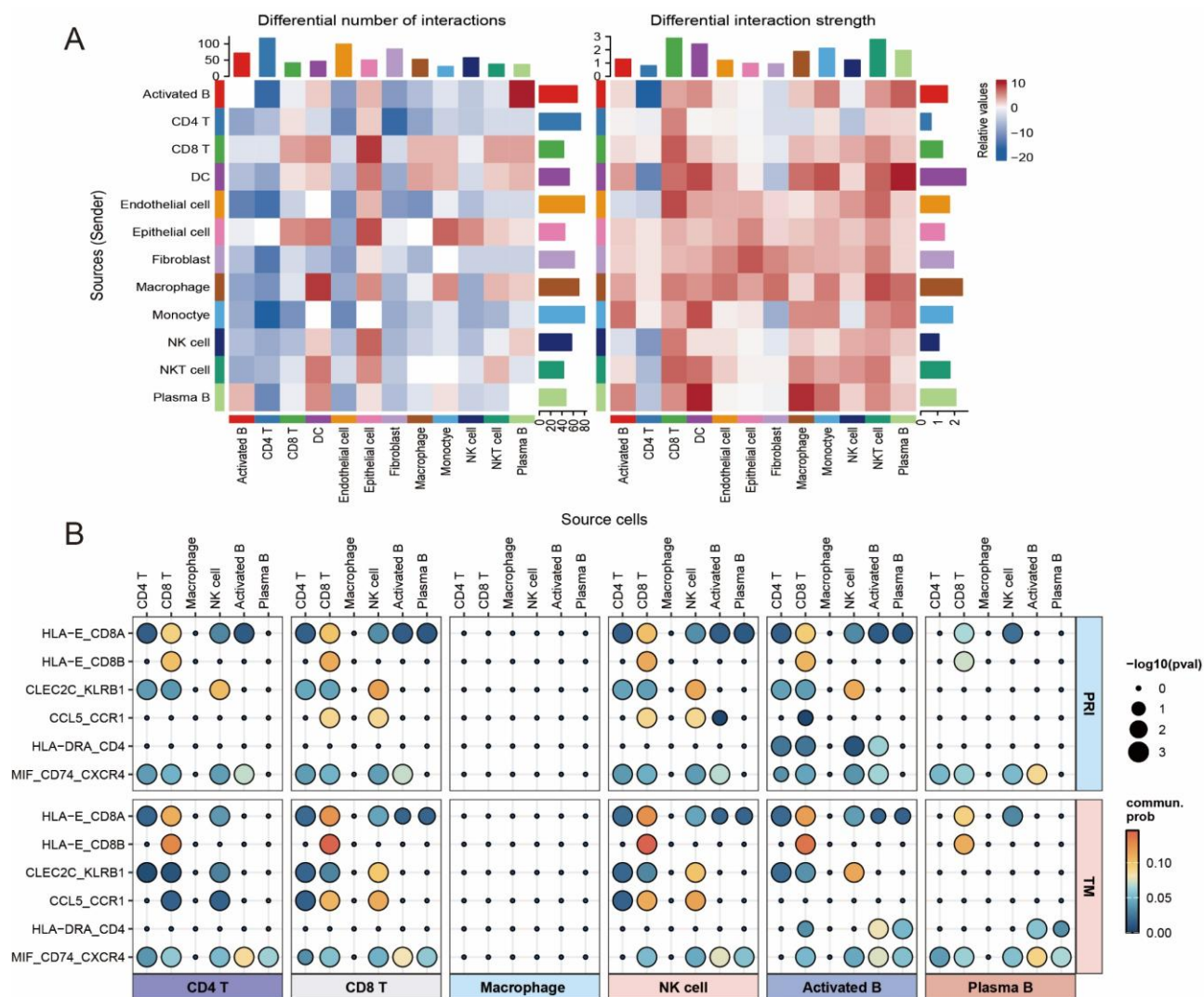
Supplementary Figure 3. Cell-cell communication landscape in the gastric cancer microenvironment.

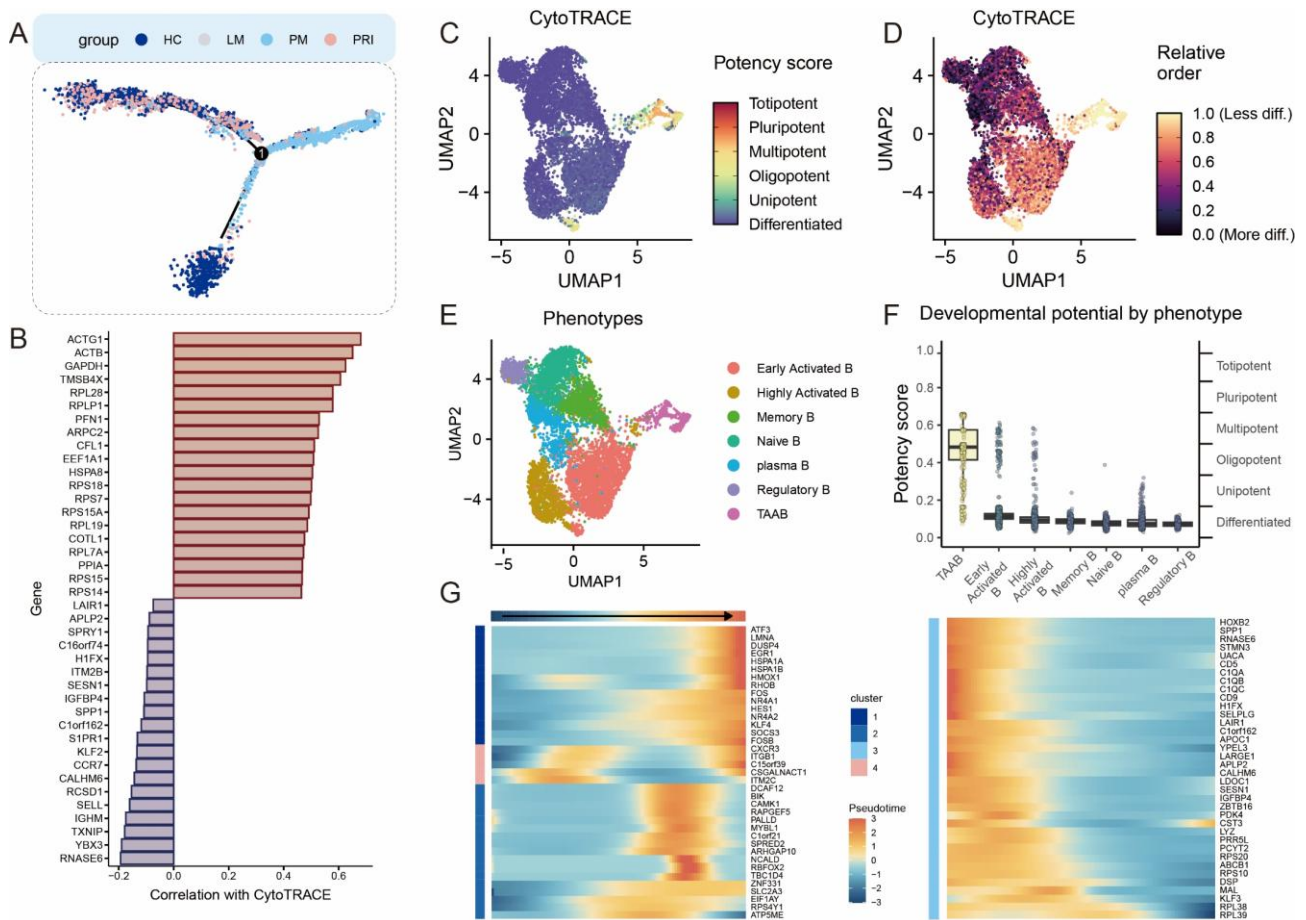
(A) Heatmaps showing the number and strength of interactions between cell types. (B) Scatter plot summarizing incoming and outgoing interaction strength for each cell type, with bubble size representing interaction count. (C) Heatmaps displaying outgoing and incoming signaling patterns for specific ligand-receptor pairs across cell populations. (D) Chord diagram illustrating the CD86 signaling pathway network, highlighting communication between immune cell subsets. (E) Sankey diagram depicting specific signaling interactions between B cell subsets (Activated B, Memory B, Naive B, Plasma B, Regulatory B, and TAAB) and other cell types including T cells, dendritic cells, epithelial cells, fibroblasts, and macrophages.



Supplementary Figure 4. Cell type distribution and functional gene clustering analysis.

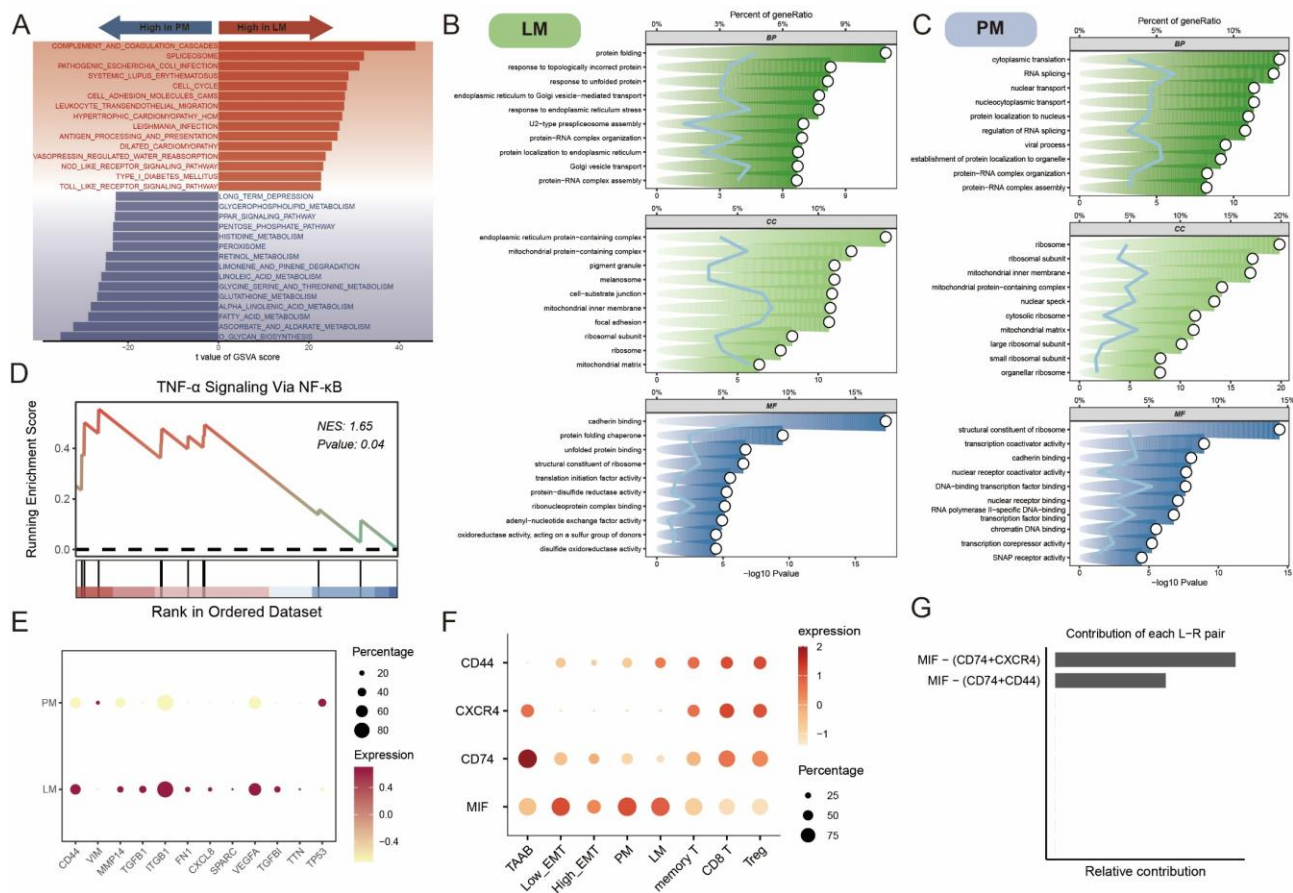
Heatmap showing cell type proportions across patient samples, including B cells, endothelial cells, epithelial cells, fibroblasts, macrophages, monocytes, NK cells, NKT cells, plasma B cells, and T cells. Nine gene clusters (C1-C9) identified through expression profiling display distinct biological processes, with representative genes labeled. UMAP visualization distinguishes normal (blue) and tumor (orange) cells based on CNV status.





Supplementary Figure 6. CytoTRACE analysis and gene expression dynamics across tissue types and B cell phenotypes.

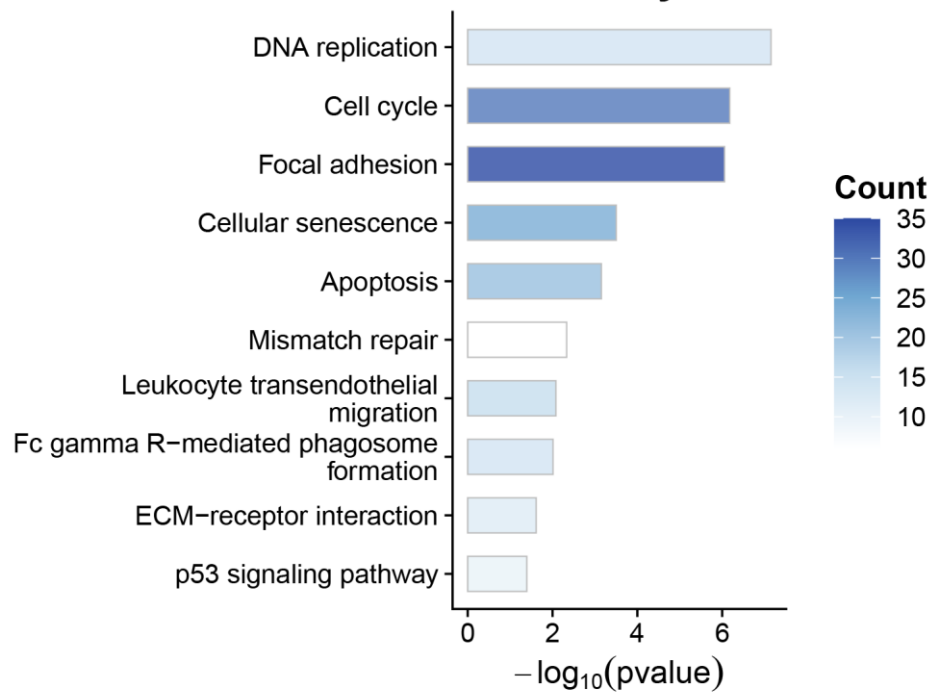
(A) UMAP visualization showing distribution of cells from different tissue sources including HC, LM, PM and PRI. (B) Bar plot displaying genes positively (brown) and negatively (purple) correlated with CytoTRACE scores, indicating their association with cellular differentiation potential. (C-D) CytoTRACE analysis mapping potency scores and relative differentiation order, with higher scores indicating stem-like properties and lower scores representing more differentiated states. (E) Distribution of B cell phenotypes including Early Activated B, Highly Activated B, Memory B, Naive B, plasma B, Regulatory B, and TAAB. (F) Box plots showing developmental potential scores across different B cell phenotypes, with TAAB exhibiting lower differentiation potential. (G) Heatmap illustrating gene expression dynamics along pseudotime, with genes clustered into four groups showing distinct temporal patterns.



Supplementary Figure 7. Pathway enrichment and ligand-receptor interaction analysis in metastatic sites.

(A) GSVA analysis showing pathways enriched in PM (red) versus LM (blue). (B-C) GO enrichment analysis for LM and PM respectively, revealing distinct biological processes, cellular components, and molecular functions. (D) GSEA plot demonstrating significant enrichment of TNF- α signaling via NF- κ B pathway (NES=1.65, P=0.04). (E) Dot plot displaying expression and percentage of key ligands across PM and LM samples. (F) Expression of MIF, CD74, CXCR4, and CD44 across different B cell subsets. (G) Bar plot showing relative contribution of MIF-(CD74+CXCR4) and MIF-(CD74+CD44) ligand-receptor pairs.

KEGG enrichment analysis



Supplementary Figure 8. KEGG pathway enrichment analysis of the turquoise module genes.