

Expanded View Figures

Figure EV1. scRNA-seq data in S2-VP10 xenografts.

- A Heatmap showing the expression of cluster-specific genes.
- B, C UMAP plots displaying selected marker gene expression.



Figure EV2. scRNA-seq data in patient PDAC.

- A UMAP plot showing the expression levels of epithelial markers.
- B Heatmap showing the expression of cluster-specific genes.
- C Gene set enrichment analysis of six subpopulations using Metascape.
- D UMAP plot displaying the expression levels of EMT-TFs.



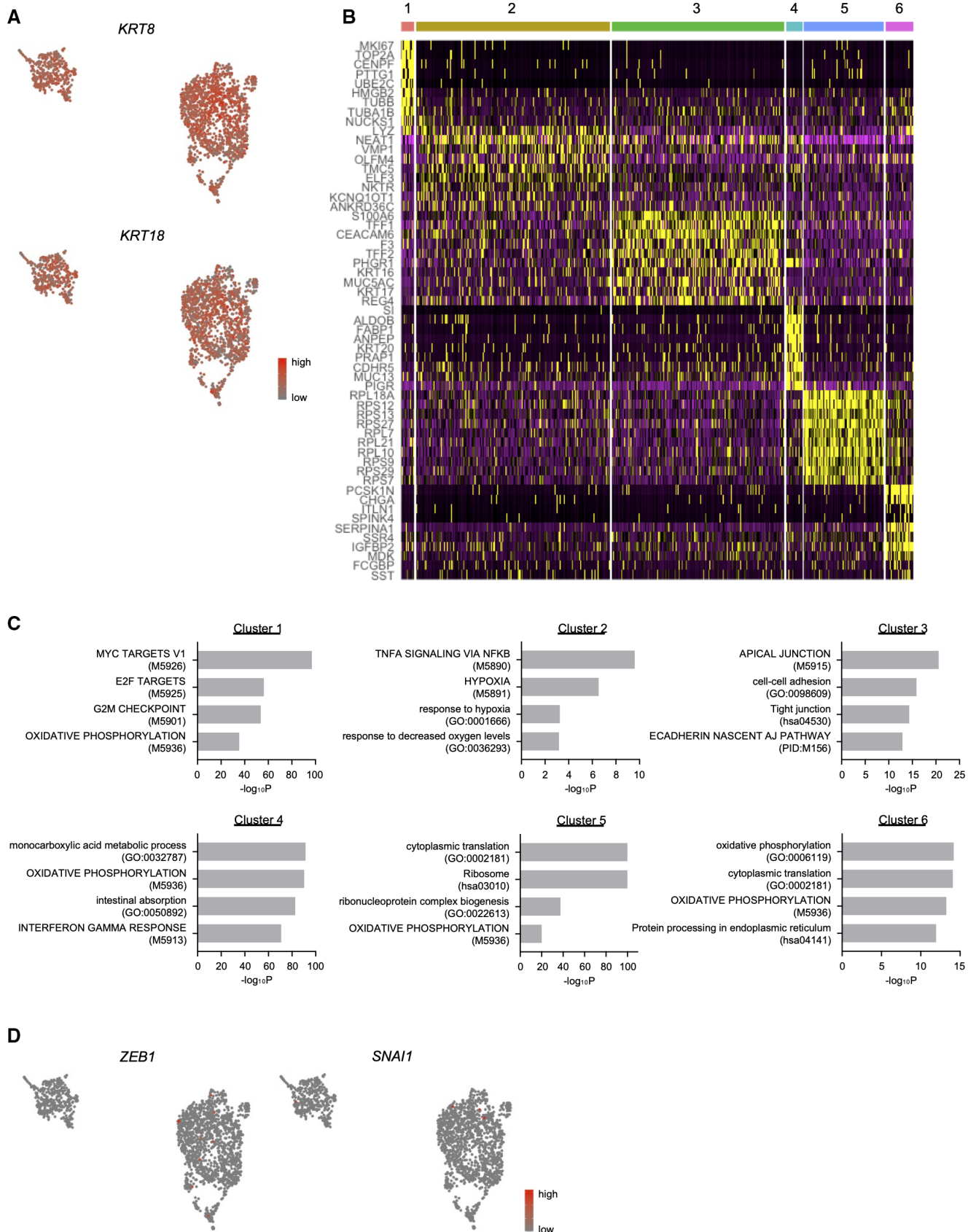


Figure EV2.

Figure EV3. Identification of the markers for partial EMT populations in PDAC.

- A Heatmap showing the expression of RTK genes within individual cells.
- B Dot plot displaying the expression of RTK genes in subpopulations.
- C Expression levels of classical EMT TF genes in ROR1^{high} cells and ROR1^{low} cells from S2-VP10 and PDO#1 xenografts ($n = 3$).
- D UMAP plots displaying known CSC marker gene expression. Dotted lines indicate the ROR1^{high} subpopulation.

Data information: Data are presented as mean \pm s.e.m., two-sided t -test. n.s., not significant.

Source data are available online for this figure.

Figure EV4. ROR1 immunostaining in patient tumors and xenografts.

A–C Representative images of patient PDAC (A), parental PDXs (B), and cell-derived xenografts (from S2-VP10 and S2-013) (C) stained with H&E and for ROR1. Black arrow, positive areas; white arrow, negative areas of ROR1.

D Representative images of Masson's trichrome and immunohistochemical staining for ROR1 and PanCK in patient-derived xenografts (PDX).

Data information: Scale bars, 100 μm (A–D).

Source data are available online for this figure.

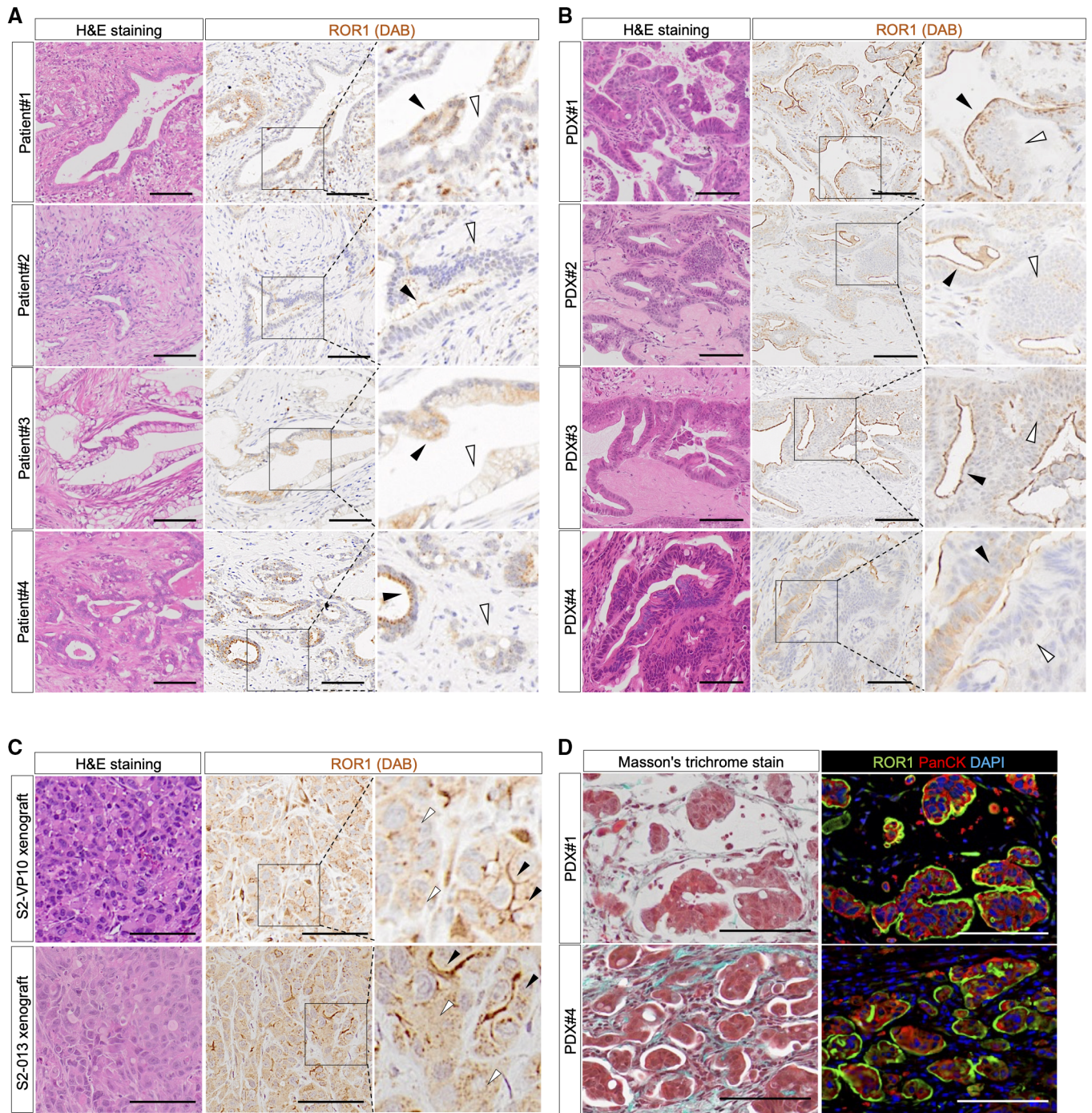


Figure EV4.

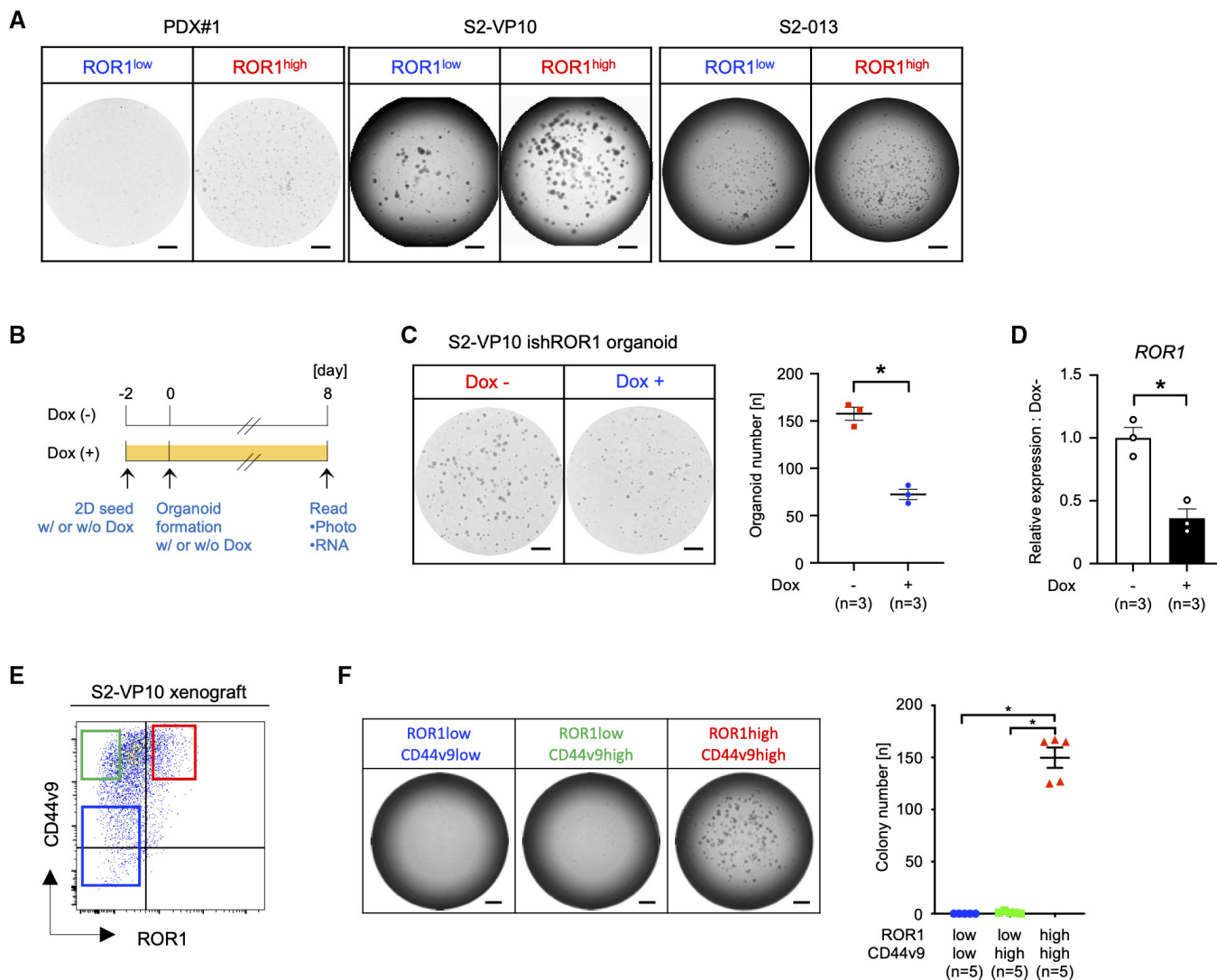


Figure EV5. Tumorigenicity of ROR1^{high} cells.

- A** Representative images of organoid or colony formation assays. The ROR1^{high} cells from PDX#1 and xenografts (S2-VP10, S2-013) efficiently formed organoids or colonies compared with ROR1^{low} cells.
- B** Schematic diagram for the evaluation of the organoid formation activity of ROR1-knockdown cells.
- C, D** Organoid formation assay of S2-VP10 ishROR1 cells with or without Dox ($n = 3$, biological replicates). Representative images, the number of organoids (C), and the expression of ROR1 (D) are shown.
- E** FACS sorting of ROR1- and CD44v9-stained cells from the S2-VP10 xenograft. Red gating: ROR1^{high}/CD44v9^{high}, green: ROR1^{low}/CD44v9^{high}, blue: ROR1^{low}/CD44v9^{low}.
- F** Colony formation assay of ROR1^{high}/CD44v9^{high}, ROR1^{low}/CD44v9^{high}, and ROR1^{low}/CD44v9^{low} cells ($n = 5$, biological replicates). Representative images of colony formation assays and the number of colonies are shown. The ROR1^{high}/CD44v9^{high} cells from S2-VP10 xenografts efficiently formed colonies.

Data information: Scale bars, 1 mm (A, C, and F). Data are presented as mean \pm s.e.m., two-sided t-test. * $P < 0.05$.

Source data are available online for this figure.

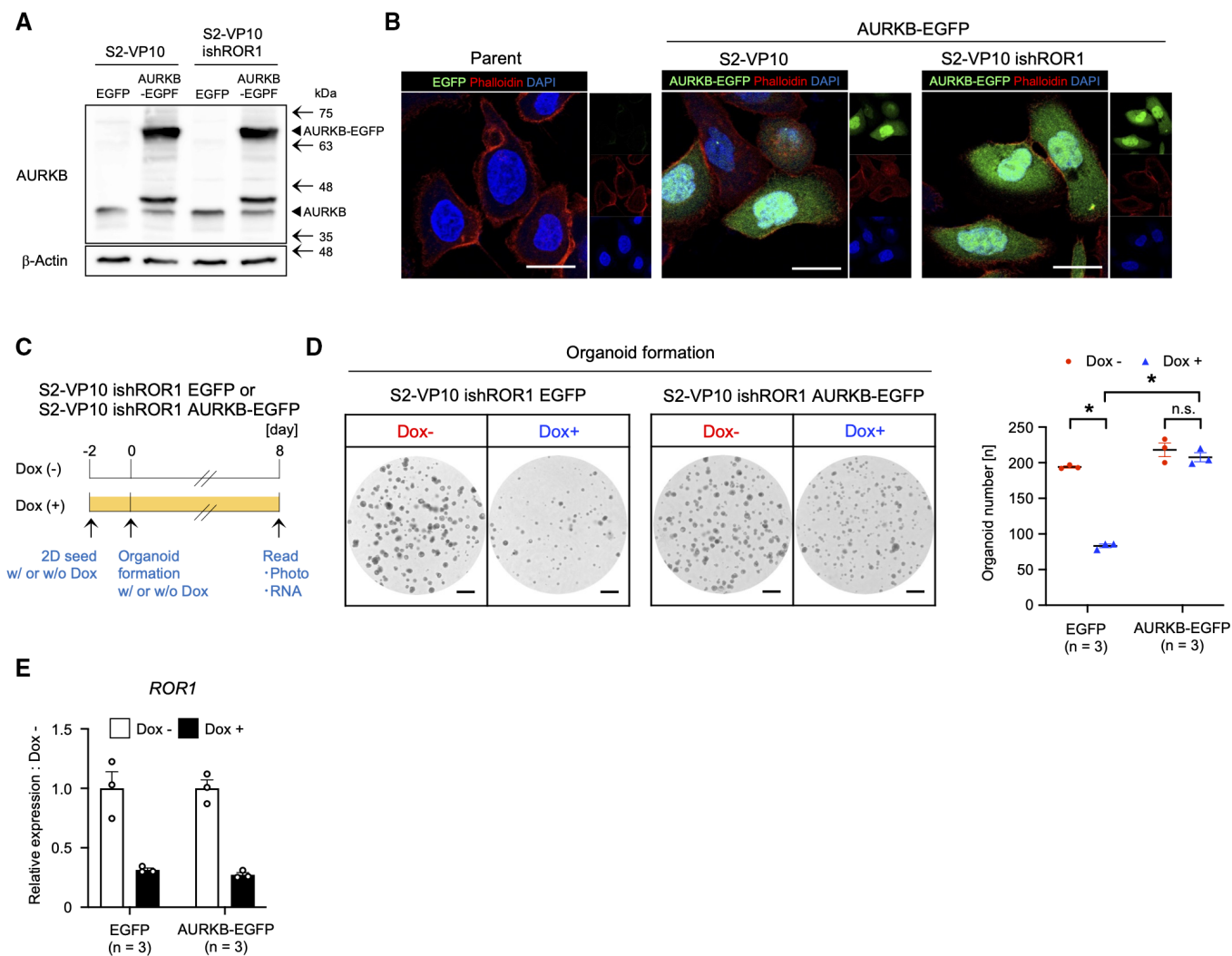


Figure EV6. Stable expression of AURKB rescues the ROR1 knockdown in organoid formation activity.

A Western blot analysis of AURKB. β-Actin was used as a loading control.
 B Cellular distribution of AURKB-EGFP protein (green) evaluated by using fluorescence confocal microscopy.
 C Schematic diagram for the evaluation of the organoid formation activity of *ROR1*-knockdown cells expressing EGFP or AURKB-EGFP.
 D, E Organoid formation assay of S2-VP10 ishROR1 cells expressing EGFP or AURKB-EGFP ($n = 3$, biological replicates). Representative images, the number of organoids (D), and the expression of *ROR1* (E) are shown.

Data information: Scale bars, 20 μm (B), 1 mm (D). Data are presented as mean ± s.e.m., two-sided t-test. * $P < 0.05$.
 Source data are available online for this figure.