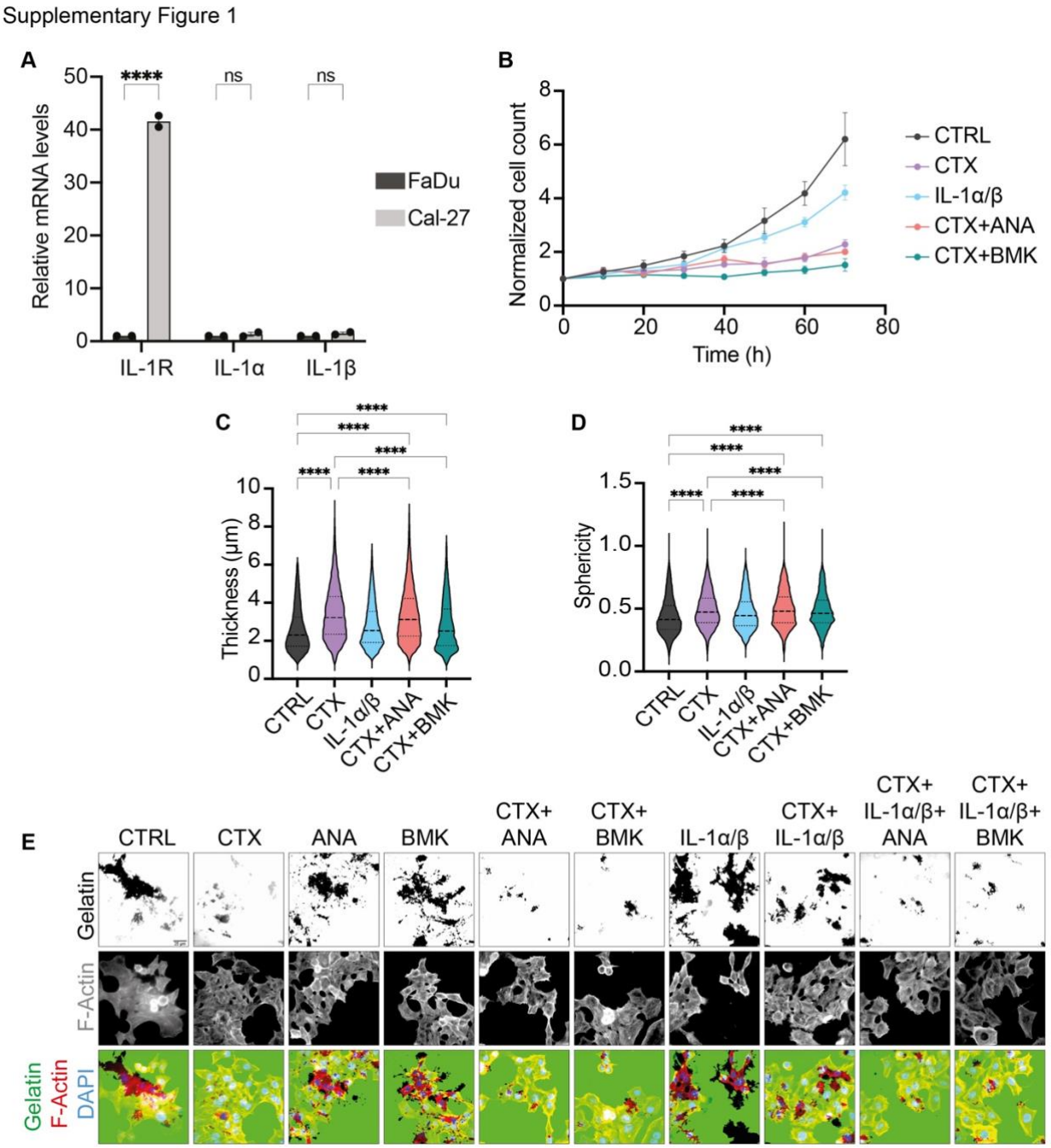
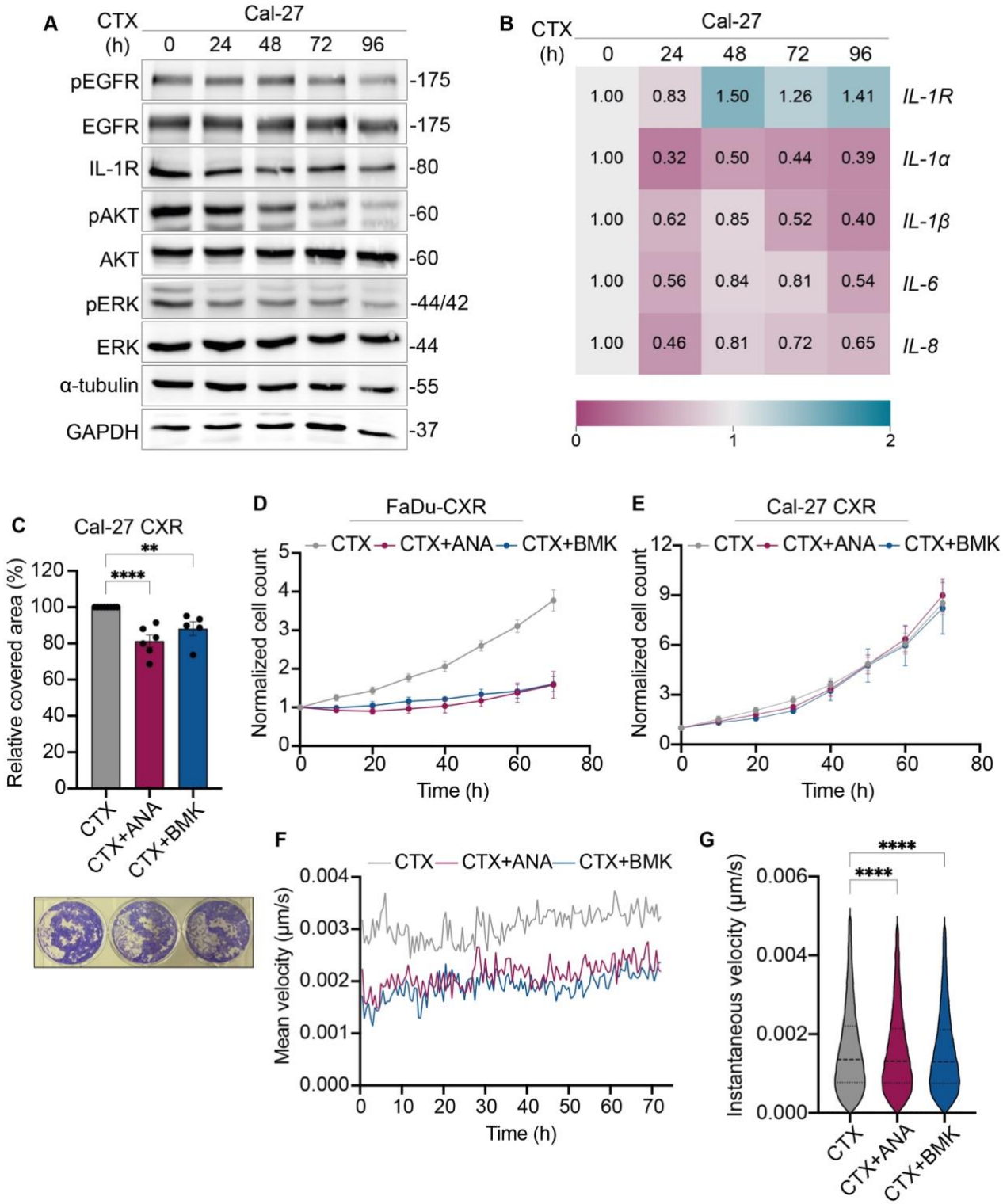
SUPPLEMENTARY FIGURES



Supplementary Figure 1. **A** IL-1R, IL-1α and IL-1β expression analysis by qPCR in FaDu and Cal-27 cell lines. B2M was used as housekeeping gene for normalization. Two-way ANOVA test was applied for statistical analysis. **B** Evaluation of FaDu cell proliferation over 72 h time-lapse performed with Phasefocus LiveCyte™. Pictures were taken every 30 min. Cells were treated with CTX (20 μg/mL), IL-1α/β (10 ng/mL each), ANA (50 μg/mL) and BMK (50 μg/mL). Data were normalized on cell count at T0. **C,D** Charts of cell thickness and sphericity from experiment in (**B**). Median and quartiles distribution are plotted. Outliers were identified and cleaned from results by means of ROUT method (Q = 1%). Statistic was calculated by one-way ANOVA test. **D** Representative images of gelatin-degradation assay performed on Cal-27 cell line in **Figure 2G**. Scale bar: 25 μm

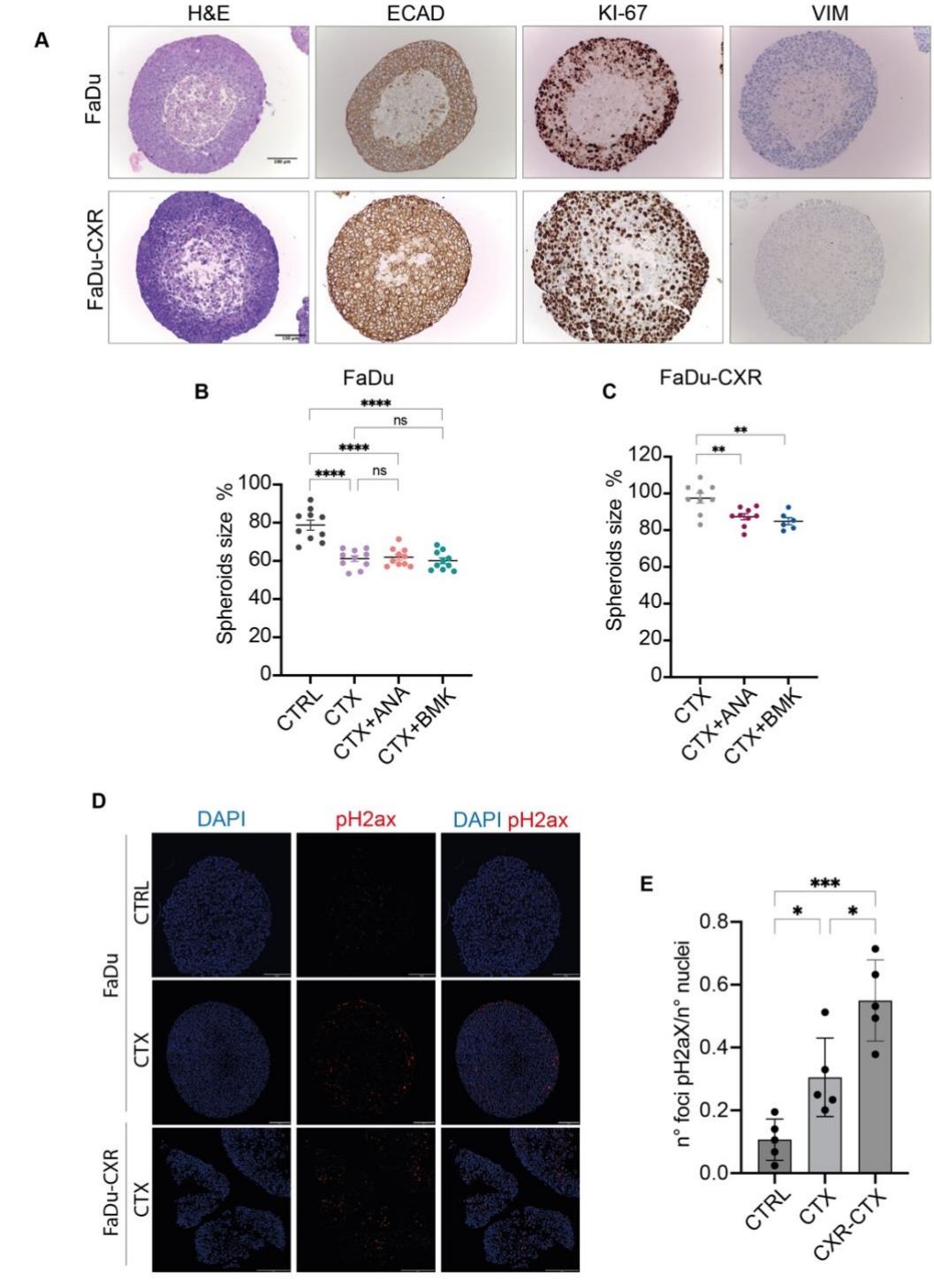
Supplementary Figure 2



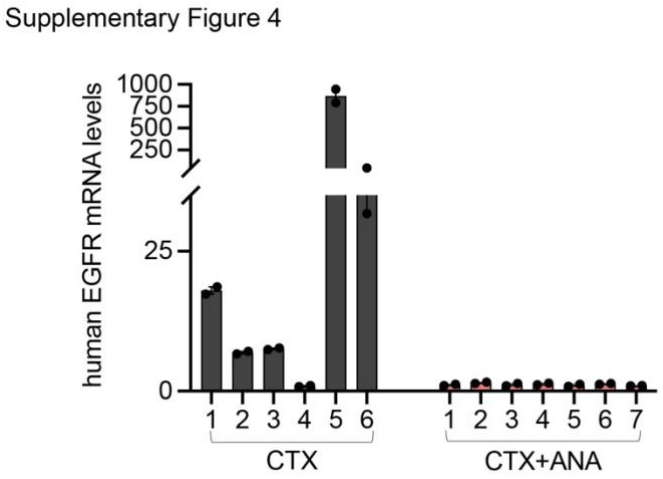
Supplementary Figure 2. A Protein analysis of EGFR signaling pathway and IL-1R in Cal-27 cells treated with CTX (50 μg/mL) over a 96 h time course. The blots for pEGFR, EGFR, pAKT and AKT belong to the same membrane and GAPDH was used as loading control. Similarly,IL-1R, pERK and ERK belong to the same membrane and α-tubulin was used as loading control. **B** Heatmap depicting mRNA levels of pro-inflammatory cytokines expression by qPCR analysis performed on Cal-27 cells treated with CTX (50 μg/mL) over a 96 h time course. B2M was used as housekeeping gene for normalization. **C** Colony forming assay of Cal-27 CXR cell line grown in CTX (50 μg/mL) and treated with ANA (50 μg/mL) and BMK (50 μg/mL). Representative image of treated wells is shown. Cell viability data are reported as % of wells covered area in relation to controls ± SEM between two

independent experiments. Statistic was calculated by one-way ANOVA test. **D** Normalized cell count of FaDu-CXR in a live-cell imaging experiment performed with Phasefocus LiveCyte™. Cells were monitored for 72 h and pictures were taken every 30 min. Cells were grown in CTX (20 μg/mL) and treated with ANA (50 μg/mL) and BMK (50 μg/mL). **E** Normalized cell count of Cal-27 CXR in a live-cell imaging experiment performed with Phasefocus LiveCyte™. Cells were monitored for 72 h and pictures were taken every 30 min. Cells were treated as in (**C**). **F** Chart of mean velocity from live-cell imaging in (**D**). **G** Chart of instantaneous velocity from live-cell imaging in (**E**). Median and quartiles distributions are plotted. Statistic was calculated by one-way ANOVA on cleaned data according to ROUT method (Q = 1%).

Supplementary Figure 3

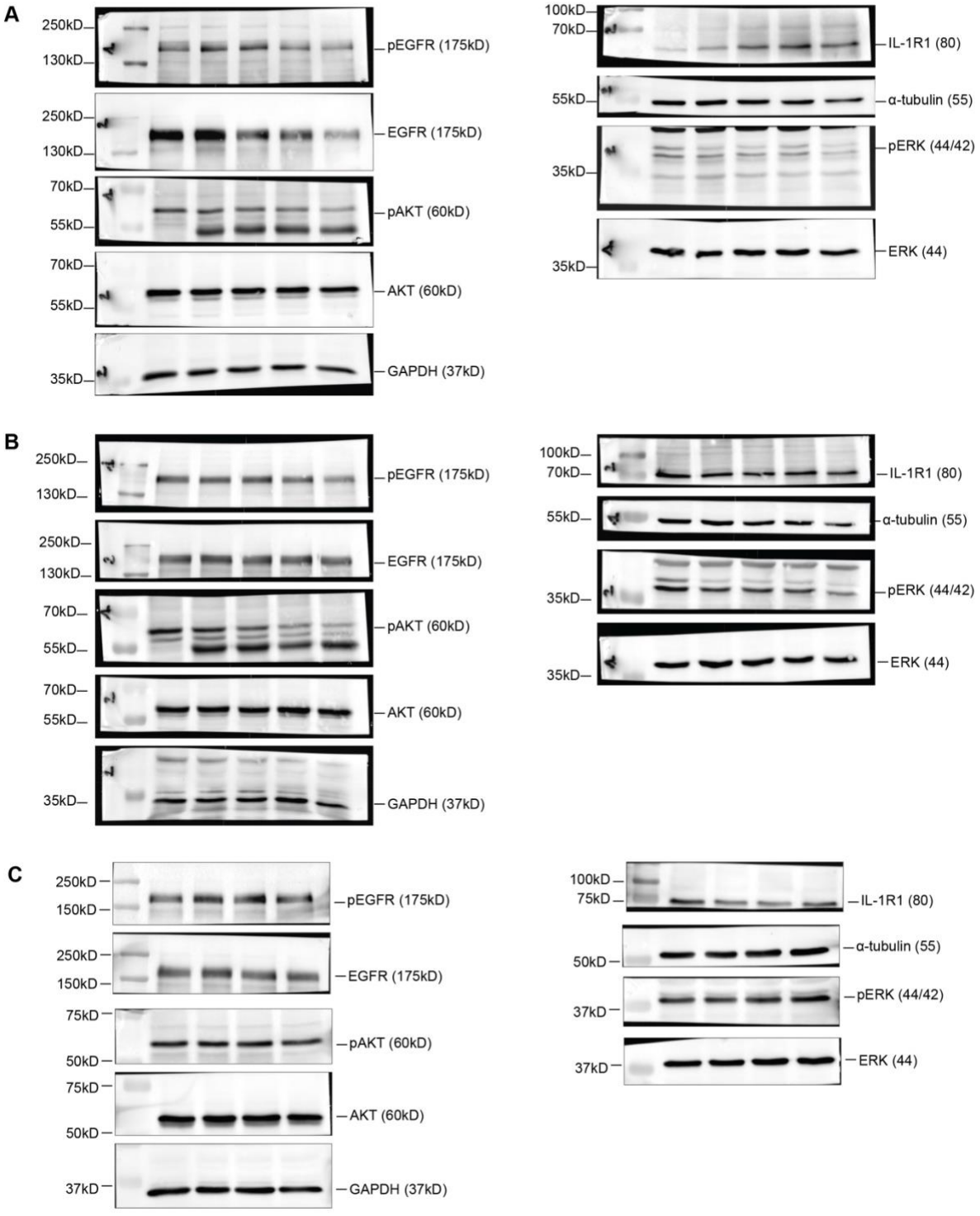


Supplementary Figure 3. A FaDu and FaDu-CXR paraffin-embedded spheroid employed for IHC assay. H&E, ECAD, Ki-67 and VIM were detected. Scale bar: 100mm. **B** Spheroid-forming assays of FaDu cell line treated with CTX (20 μg/mL), IL-1α/β (10 ng/mL each), ANA (50 μg/mL) and BMK (50 μg/mL) for 96 h. One-Way ANOVA was applied for statistical analysis. **C** Spheroid-forming assays of FaDu-CXR cell line grown in CTX (20 mg/mL) and treated with ANA (50 mg/mL) and BMK (50 mg/mL) for 96 h. Statistic was calculated by one-way ANOVA test. **D, E** Immunofluorescence assay on FaDu and FaDu-CXR paraffin-embedded spheroids. FaDu spheroids were treated with CTX (20 mg/mL) for 96 h and FaDu-CXR spheroids were grown in CTX (20 mg/mL). Spheroids were stained with pH2ax (red) and DAPI (nuclei) prior to visualization. Representative images were acquired by scanner Olympus VS200, scale bar: 200 μm (**D**). Quantification was performed on at least 5 spheroids per condition and foci count was normalized over the nuclei of each spheroid (**E**). Data are reported as mean ± SEM. Statistic was calculated by One-way ANOVA.



Supplementary Figure 4. Determination of human EGFR by qPCR analysis performed on mouse lungs from *in vivo* experiment. Mouse GAPDH was used as housekeeping gene for normalization. Mice were previously tail vein injected with FaDu-CXR cells grown in CTX (20 μg/mL) and then treated with CTX alone (100 mg/injection) or in combination with ANA (10 mg/kg) for 4 weeks.

Supplementary Figure 5



Supplementary Figure 5. A Raw membranes merged with matching loading control of western blot in Figure 3A. **B** Raw membranes merged with matching loading control of western blot in Supplementary Figure 2A. **C** Raw membranes merged with matching loading control of western blot in Figure 5D.