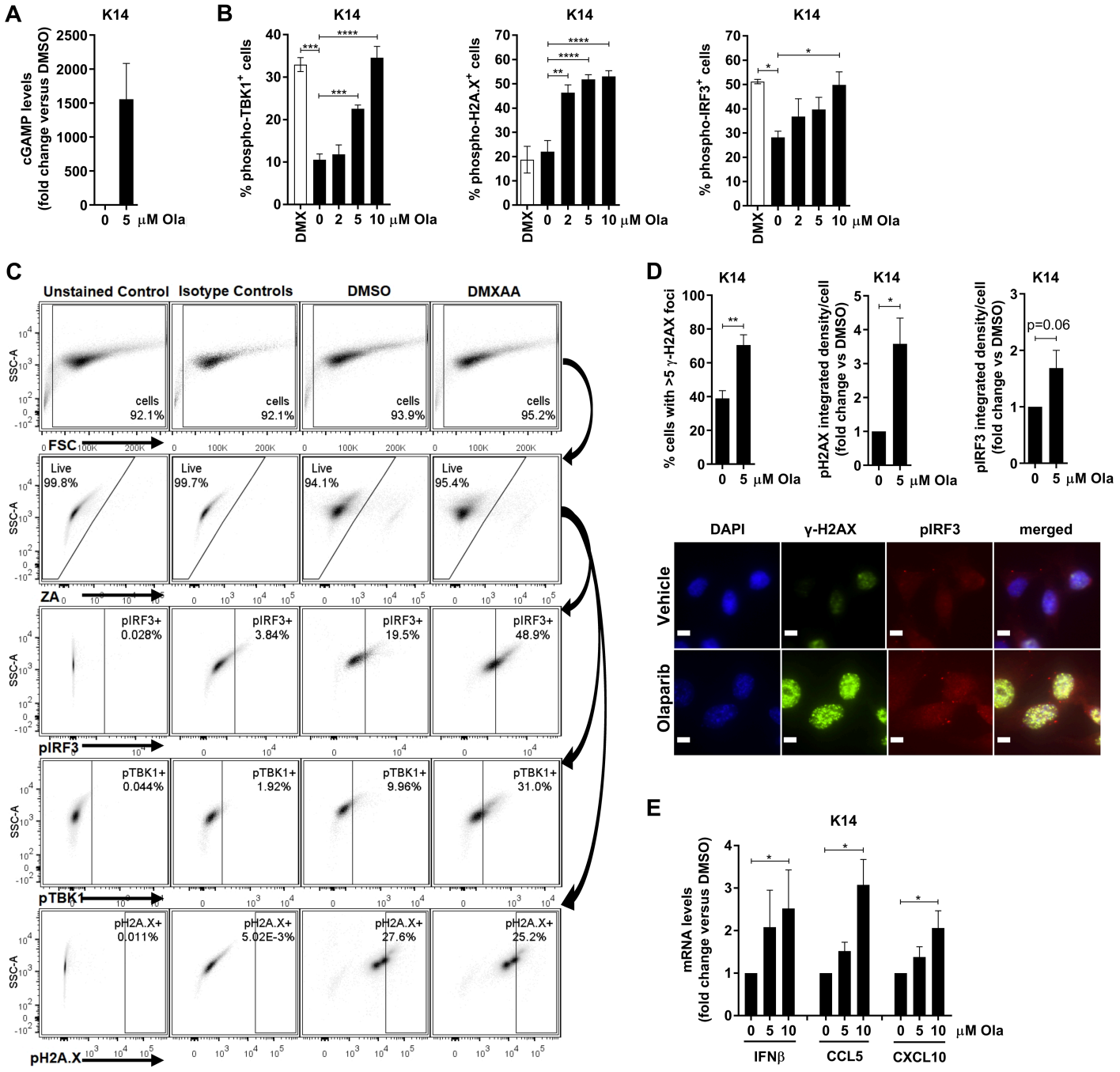

Supplementary Figure S3

(A) The murine K14-Cre BRCA1$^{f/f}$p53$^{f/f}$ cell line (K14) was treated with DMSO (0 μM) or the indicated doses of olaparib for 72h and subjected to ELISA for cGAMP production. Error bars represent SEM of 3 independent experiments. (B) K14 cells were treated with the indicated doses of Olaparib for 72h and subjected to flow cytometric analysis of pTBK1^{Ser172}, pIRF3^{Ser396} and pH2AX^{Ser139} expression. DMXAA (10 μM, 24h) was used as a positive control. Error bars represent SEM of 2-4 independent experiments. Statistical analyses were performed using one-way ANOVA with Sidak's post-hoc test. (C) Gating strategy used in flow cytometric analysis of pIRF3, pTBK1 and pH2AX in cell lines. Debris was excluded and zombie-aqua positive i.e. non-viable cells were gated out. Live cells were analyzed for pIRF3, pTBK1 and pH2AX expression. The examples shown are from unstained control, isotype controls, vehicle- and DMXAA-treated K14 cells. (D) K14 cells were treated with 0 or 5 μM olaparib for 24h and subjected to immunofluorescence staining for pH2AX and pIRF3. The number of cells displaying >5 γ-H2AX foci was quantified, and statistical analysis was performed using unpaired t-test. pIRF3 and pH2AX corrected integrated density/cell was expressed as fold change versus DMSO. Statistical analyses were performed using one-sample t-test. Error bars represent SEM of 3 independent experiments. Representative images of DAPI (blue), γ-H2AX (green) and pIRF3 (red) stained cells are shown (20x magnification) on the bottom panel. Scale bar, 8 μm. (E) K14 cells were treated with the indicated doses of olaparib for 72h and analyzed for mRNA expression of IFNβ, CCL5 and CXCL10 (normalized to GAPDH internal control). Error bars represent SEM of 4 independent experiments. Statistical analysis was performed using Kruskal-Wallis test Dunn's post-hoc test.