

Expanded View Figures

Figure EV1. High *ESRP2* expression is not associated with prognosis in both ER+ and ER– breast tumors.

- A BreastMark microarray platform; Kaplan–Meier curves of overall survival (OS) in ER-positive (ER+) breast cancer. A log rank test was used to calculate the $P = 0.47$ ($n = 708$, number of events = 171) and $P = 0.13$ ($n = 253$, number of events = 96).
- B BreastMark microarray platform; Kaplan–Meier curves of overall survival (OS) in ER-negative (ER–) breast cancer. A log rank test was used to calculate the $P = 0.13$ ($n = 253$, number of events = 96).
- C TCGA Breast Invasive Carcinoma (BRCA) RNA-Seq dataset; Kaplan–Meier curves of overall survival (OS) in ER+ breast cancer. A log rank test was used to calculate the $P = 0.45$ ($n = 210$, number of events = 49).
- D TCGA Breast Invasive Carcinoma (BRCA) RNA-Seq dataset; Kaplan–Meier curves of overall survival (OS) in ER– breast cancer. Red, high *ESRP1* expression; black, low *ESRP1* expression. A log rank test was used to calculate the $P = 0.24$ ($n = 129$, number of events = 21).
- E *ESRP1* expression in breast cancer with poor prognosis. RT–qPCR assay showing that expression of *ESRP1* is higher in patients with high recurrence scores in a Oncotype DX cohort $n = 59$ (19 LS, 20 IS, and 20 HS); LS, low score; IS, intermediate score; and HS, high score, * represents $P < 0.05$; data (mean \pm SD) were statistically significant and calculated using two-way ANOVA.

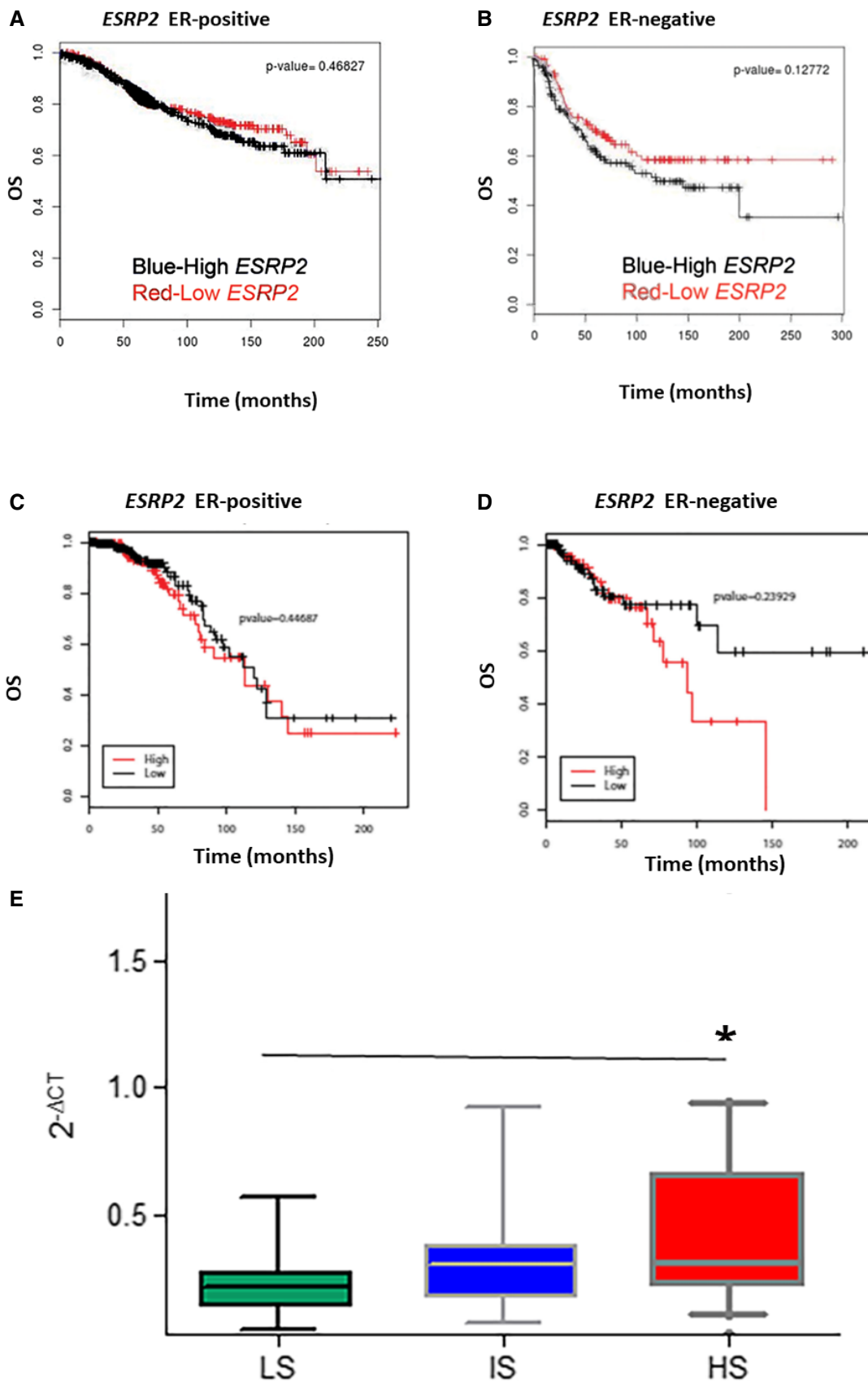


Figure EV1.

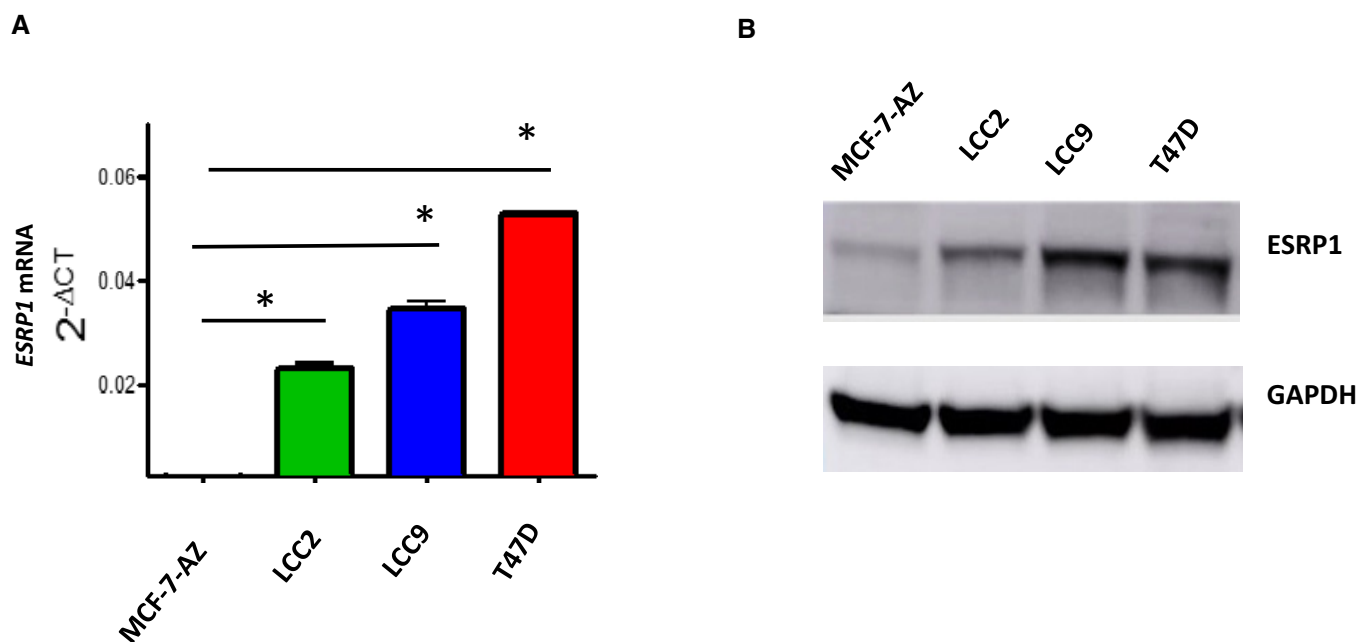


Figure EV2. ESRP1 expression in ER+ breast cancer cell lines at the mRNA and protein levels.

A RT-qPCR assay showing *ESRP1* expression in endocrine-resistant (MCF7/LCC2-tamoxifen resistant, MCF7/LCC9-fulvestrant and tamoxifen cross-resistant) and T-47D cells compared to parental MCF-7-AZ therapy-sensitive cell lines. * represents $P < 0.05$; statistically significant. All RT-qPCR assays are performed in three independent biological replicates calculated using two-way ANOVA (data (mean \pm SD)).

B Representative Western blot analysis of the indicated breast cancer cell lines (see details in Materials and Methods). All Western blot assays are performed in three independent biological replicates.

Source data are available online for this figure.

Figure EV3. Agilent Seahorse XFp Cell Energy Phenotype Assays in *ESRP1* knockdown cells compared to their control resistant cell lines using Agilent Seahorse Cell XF Technology.

- A Agilent Seahorse XF Cell Energy Phenotype Profile. The relative utilization of the two energy pathways of control and *ESRP1* knockdown cells is determined under both baseline (Baseline Phenotype-open squares) and stressed (Stressed Phenotype-full squares) conditions—2-control (blue squares) versus 2C3 (red squares).
- B Agilent Seahorse XF Cell Energy Phenotype Profile. The relative utilization of the two energy pathways of control and *ESRP1* knockdown cells is determined under both baseline (Baseline Phenotype-open squares) and stressed (Stressed Phenotype-full squares) conditions—9-control (blue squares) versus 9C2 (red squares).
- C Agilent Seahorse XF Glycolysis Stress representing extracellular acidification rate (ECAR) components—glycolysis, glycolytic capacity, and glycolytic reserve—2-control (green columns) and -2C3 *ESRP1* knockdown (blue columns). Data (mean \pm SD) are calculated using two-way ANOVA based on the three independent biological replicates.
- D Agilent Seahorse XF Glycolysis Stress representing extracellular acidification rate (ECAR) components—glycolysis, glycolytic capacity, and glycolytic reserve—9-control (light blue columns) and 9C2 *ESRP1* knockdown (orange columns). Data (mean \pm SD) are calculated using two-way ANOVA based on the three independent biological replicates.
- E Mitochondrial respiration through oxygen consumption rate (OCR) components—basal respiration and spare respiration capacity. The data are representative of three independent sets. * represents $P < 0.05$; statistically significant. Data (mean \pm SD) are calculated using two-way ANOVA based on the three independent assays.

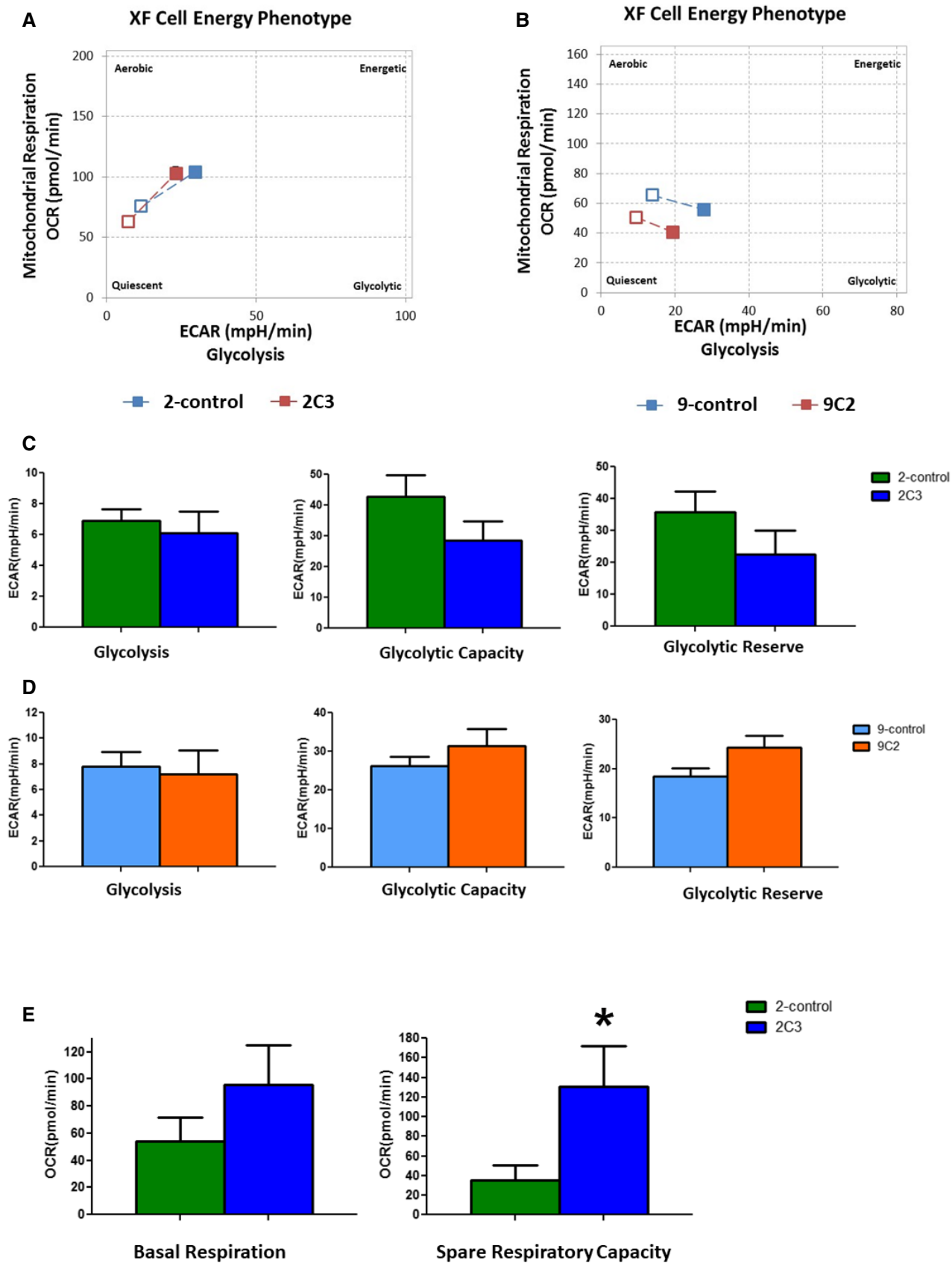


Figure EV3.