

Figure S1. CD44 and CD24 expression in AI-sensitive and AI-resistant T47D cells. (A) CD44 and (B) CD24 expression in aromatase-transfected T47D letrozole-sensitive cells (T47Darom) and T47Darom letrozole-resistant breast cancer cells (T47DaromLR), cultured adherently or as mammospheres. All cells were assayed by immunoblotting to examine the expression levels of CD24, CD44 and GAPDH (loading control). (C) Representative immunoblots showing the protein expression levels of CD24, CD44 and GAPDH. The immunoblot images are representative of more than three independent experiments with a minimum of two duplicates per sample. Graphs depict the percentages of protein expression relative to 2D cell counterparts. Comparison of 2D vs. 3D treatments were analyzed by Student's t-test using GraphPad Prism software. *** $P < 0.0004$, ** $P < 0.0024$, * $P \leq 0.038$. AI, aromatase inhibitor.

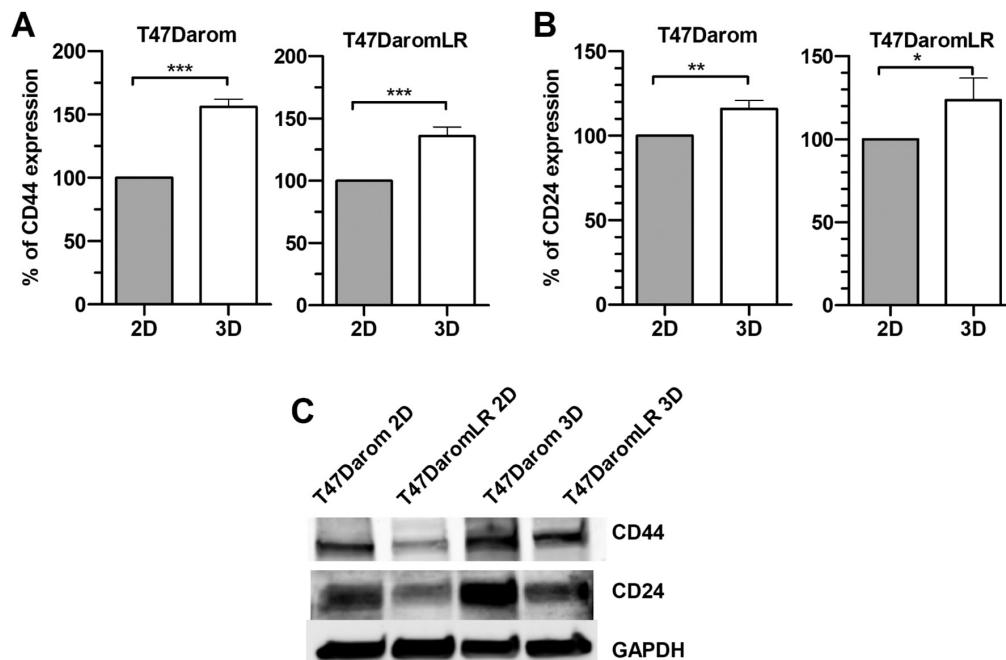


Figure S2. Growth of AC-1 and LTLT-Ca tumors in xenograft mice. AC-1 and LTLT-Ca cells were injected into the mammary fat pad (n=5 for each group). For mice containing AC-1 cells, estrogen pellets were subcutaneously implanted. Tumors were allowed to form over 10 days. Tumor volume was measured weekly for 8 weeks using a digital caliper and the tumor volume was calculated using the following formula: $4/3LM^2$, where L is the larger radius and M is the smaller radius. Values indicate tumors grouped from AC-1 and LTLT-Ca xenografts.

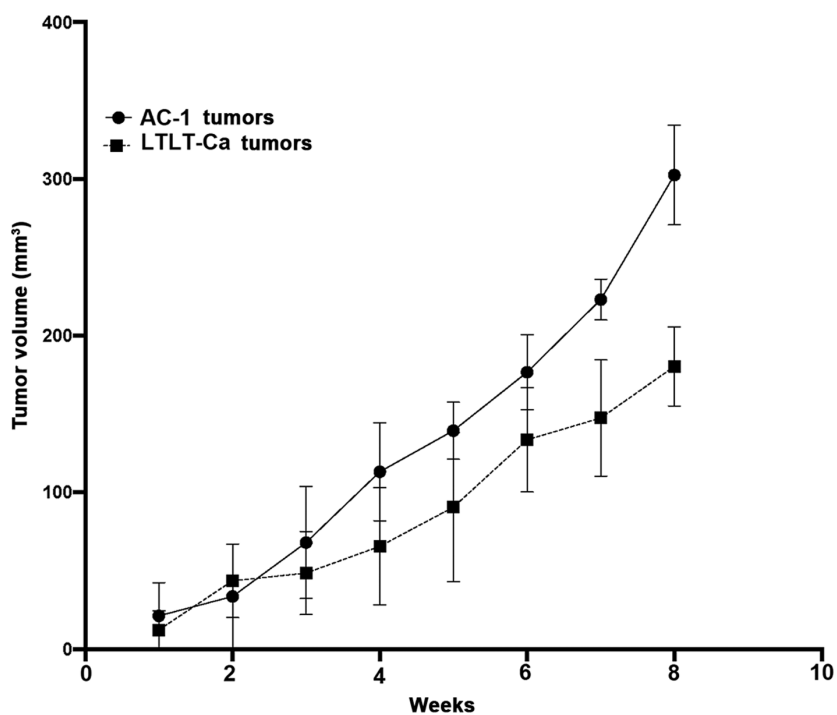


Figure S3. LTLT-Ca mammospheres were formed and representative images were stained with DAPI (blue) in the top panels, anti-CD24 (green), anti-CD44 (green) or anti-Ki67 (red) in the middle panels. In the bottom panels, images were merged and captured using an Olympus BX41 microscope. Original magnification, x100. Cells were fluorescently labeled with either TRITC or FITC. FITC, Fluorescein isothiocyanate; TRITC, tetramethylrhodamine.

