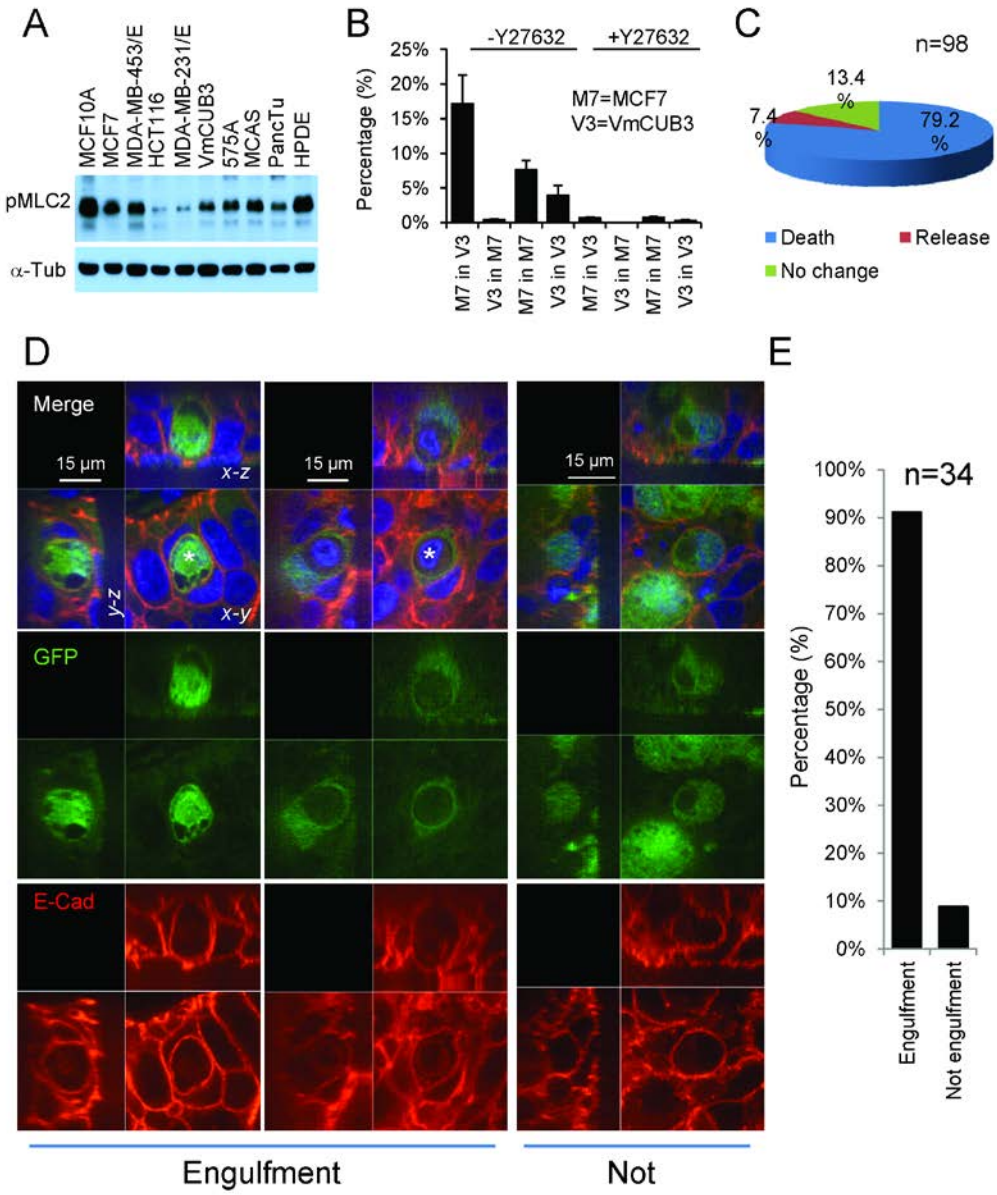

Figure S1

Supplementary information, Figure S1 Entosis-mediated cell competition in vitro and in vivo. Related to Figure 1.

(A) Expression level of pMLC2 across a panel of different human cell lines. Cells were cultured in suspension for 3 hours and then lysed for western blotting. MCF10A (mammary) and HPDE (pancreatic) are non-transformed human epithelial cells, the remaining cell lines are human tumor cells: MCF7 is breast carcinoma, MDA-MB-453/E and MDA-MB-231/E are breast carcinoma expressing exogenous E-cadherin, 575A and VmCUB3 are bladder carcinoma cell lines, HCT116 is colon carcinoma, MCAS is ovarian carcinoma, and PancTu is pancreatic carcinoma.

(B) Cell-in-cell structure formation of VmCUB3/MCF7 co-culture in suspension for 7 hr with and without Y27632, an inhibitor of entosis. Data are mean±sd of three experiments. n=903 cells.

(C) Internalized cell (MCF7) fates measured by time-lapse analysis of entotic cell structures of MCF7 inside of VmCUB3. Data are means of an experiment performed in triplicate, n=98 entotic structures.

(D) Sections of 30 μm were immunostained for E-cadherin (red) and GFP (green) to image cell engulfment by confocal microscopy. Representative images for two engulfments (left panels) and a cell that is not engulfed (right panel). Asterisks indicate inner cells.

(E) Quantification of cell engulfment in thick xenograft tumor sections. For quantification, individual cell engulfments were identified at the midplane, and then analyzed through entire z-stack. As shown in the graph, 91% of structures were engulfments when imaged through all z-planes, and only 9% were not cell engulfments, as less than 2/3 of the cell body was internalized when examined through all z-planes. n=34.