Supplemental Figure 1. *Par6 overexpression does not alter late stage acinar organization.* Immunofluorescence staining of Day 20 acini structures grown in 0.5 ng/ml EGF with the basal marker Laminin V (Red) and co-stained with DAPI to monitor nuclei. Scale bar represents 20 μm.

Supplemental Figure 2. *Par6β overexpression promotes proliferation in 2D, 3D and activates MAPK* **(A)** *Flag-Par6β is overexpressed in MCF-10A cells.* Cell extract from vector control and Par6β expressing cells were immunoblotted with anti-flag and reprobed with anti-β-actin antibodies. **(B)** *Par6β overexpression promotes EGF-independentt cell proliferation.* Cell cycle analysis of cells grown for 3 days in EGF free media. Data are fold increase in S-phase of Par6β cells compared to control cells and are means +/- s.d. of three independent experiments. **(C)** *Par6β overexpression promotes acini cell proliferation.* Distribution of acini size (circumferential area) of Day 12 structures grown in 0.5ng/ml EGF. Each condition represents approximately 800 acini structures from three independent experiments. The P-value between Vector and Par6β is less than 0.0001 calculated by Wilcoxon rank-sum test. **(D)** *Par6β overexpression activates MAPK signaling.* Cell extracts that were stimulated for 0,15, 60 and

120 minutes with 2 ng/ml EGF were immunoblotted with antibodies specific for phosphorylated ERK1/2 and reprobed with total ERK2.

Supplemental Figure 3. *Par6 overexpression increases the cell number/acini*. **(A)** Distribution of nuclei per acini structure. Day four acini structures grown in 0.5 ng/ml EGF were stained with DAPI to monitor nuclei. Each nuclei within each acini was counted. The data represents approximately 300 acini.

Supplemental Figure 4. *Par6 overespression promotes hyperplastic acinar structures and does not alter apoptosis in acinar structures.* (**A**) *Par6 overespression promotes proliferation in late stage acinar structures.* Quantitation of Day 16 and Day 20 acini with at least one positive Ki-67 positive nuclei within the acinar structure. Acini were grown in 0.5ng/ml EGF. Data for Day 16 are represented as percent Ki-67 positive structures +/- s.d. Each bar represents approximately 700 acini from three independent experiments. Data from Day 20 are represented as percent Ki-67 positive acinar structures from approximately 200 acini from one experiment. (**B***) Par6 expression does not alter the onset of cell death during acinar morphogenesis.* Day 7 acinar structures derived from Par6 expressing or vector control cells were incubated with ethidium bromide to mark the cells that underwent cell death during acinar morphogenesis. Scale bar represents 100 μm. (**C**) *Par6 overexpression does not promote apoptosis in late stage acinar structures.* Immunofluorescence staining of Day 20 acini structures grown in 0.5 ng/ml EGF with the apoptotic marker cleaved-caspase-3 (Green) and co-stained with DAPI to monitor nuclei. The lack of cleaved-caspase-3 staining demonstrates that as expected there is no apoptosis within the lumen of the acinar structures. Scale bar represents 50 μm.

Supplemental Figure 5. *Par6 promotes proliferation of mammary epithelial cells.*

(A) Phase contrast images showing growth inhibited vector control MCF-10A cells compared to growing Par6α overexpresssing cells in EGF free media. The scale bar represents 100 μm. **(B)** Cell cycle analysis of cells grown for 3 days in EGF free media. Data are fold increase in S-phase of Par6 cells compared to control cells and are means +/- s.d. of three independent experiments. **(C)** Phase images showing growth-inhibited vector control Comma1D cells compared to growing Par6α overexpresssing in EGF free media. The scale bar represents 100 μm. **(D)** Proliferation analyses of cells grown for three days in EGF free media by estimating the cell number with a hemocytometer. Data are means +/- s.d.from three independent experiments. **(E)** Phase contrast images of larger Par6α overexpressing HC11 acini structures compared to vector control grown in 0.5 ng/ml EGF for 12 day. Scale bar represents 100 μm. **(F)** Par6 overexpression significantly increases HC11 acini size compared to vector control. Distribution of acini size (circumferential area) of day 12 HC11 acini structures grown in 0.5 ng/ml EGF. Data is plotted as a box plot and represents approximately 200 acini (P=0.03).

Supplemental Figure 6. *Par6 mutations do not activate MAPK.* MCF-10A cell extracts that were stimulated for 0 and 60 minutes with 2 ng/ml EGF were immunoblotted with antibodies specific for phosphorylated ERK1/2 and reprobed with total ERK2.

Supplemental Figure 7. *Pard6b is overexpressed in ER positive tumors.*

Meta-analysis of published databases using Oncomine an integrated data platform (31). **(A)** Analysis of 69 ER^- and 226 ER^+ breast carcinoma, P-value = 2.4E-23 (33). **(B)** Analysis of 11

ER^- and 24 ER^+ breast carcinoma, P-value = 4.6E-7 (34). **(C)** Analysis of 28 ER^- and 27 ER^+ breast carcinoma, P-value = 9.5E-7(23). **(D)** Analysis of 9 ER^- and 26 ER^+ breast carcinoma, P-value = 7.1E-4 (32).

Supplemental Figure 8. Tamoxifen does not regulate *Pard6b levels in MCF-7 cells.*

Quantitative PCR analysis of Par6β gene expression using cDNA generated from MCF-7 cells. Data is represented as fold increase over MCF-7 untreated control after normalization to b-actin gene expression. MCF-7 cells were grown for 3 days in media containing charcoal treated fetal bovine serum and treated with tamoxifen for 3 hours before RNA extraction.