

Supplemental Figure S1: J.R. Neil and W.P. Schiemann

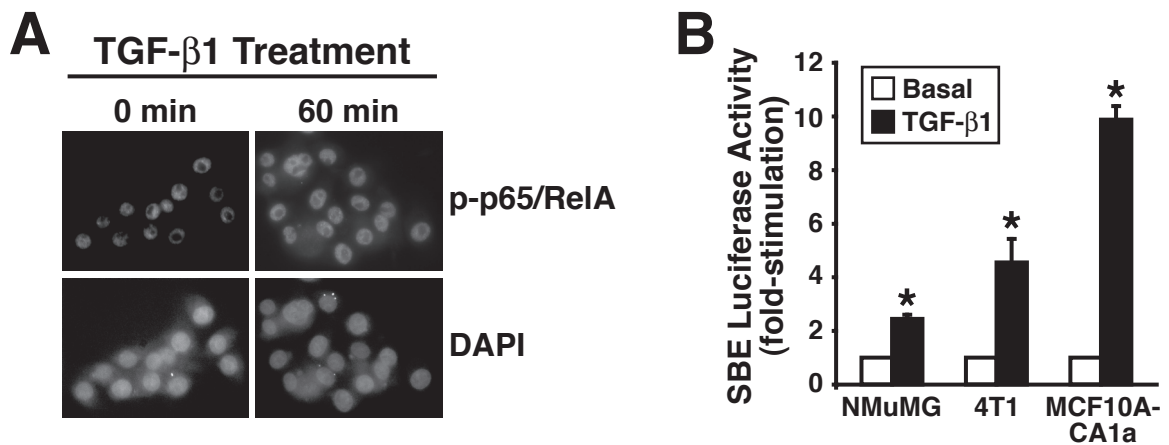


Figure S1. Response of normal and malignant breast cancer cells to TGF- β . (A) Human MCF10A-CA1a breast cancer cells were stimulated with TGF- β 1 (5 ng/ml) for 60 min. Afterward, the cells were fixed in 4% paraformaldehyde and processed for phosph-p65/RelA immunofluorescence as described previously (33). Photomicrographs are from a representative experiment that was repeated twice with identical results. (B) NMuMG, 4T1, and MCF10A-CA1a cells were transiently transfected with SBE-luciferase and β -gal, and subsequently were stimulated overnight with TGF- β 1 (5 ng/ml) prior to measuring luciferase and β -gal activities contained in detergent-solubilized whole cell extracts. Data are the mean (\pm SE; n=3) luciferase activities relative to unstimulated cells. (*, $P < 0.05$; Student's T-Test).

Supplementary Figure S2: J.R. Neil and W.P. Schiemann

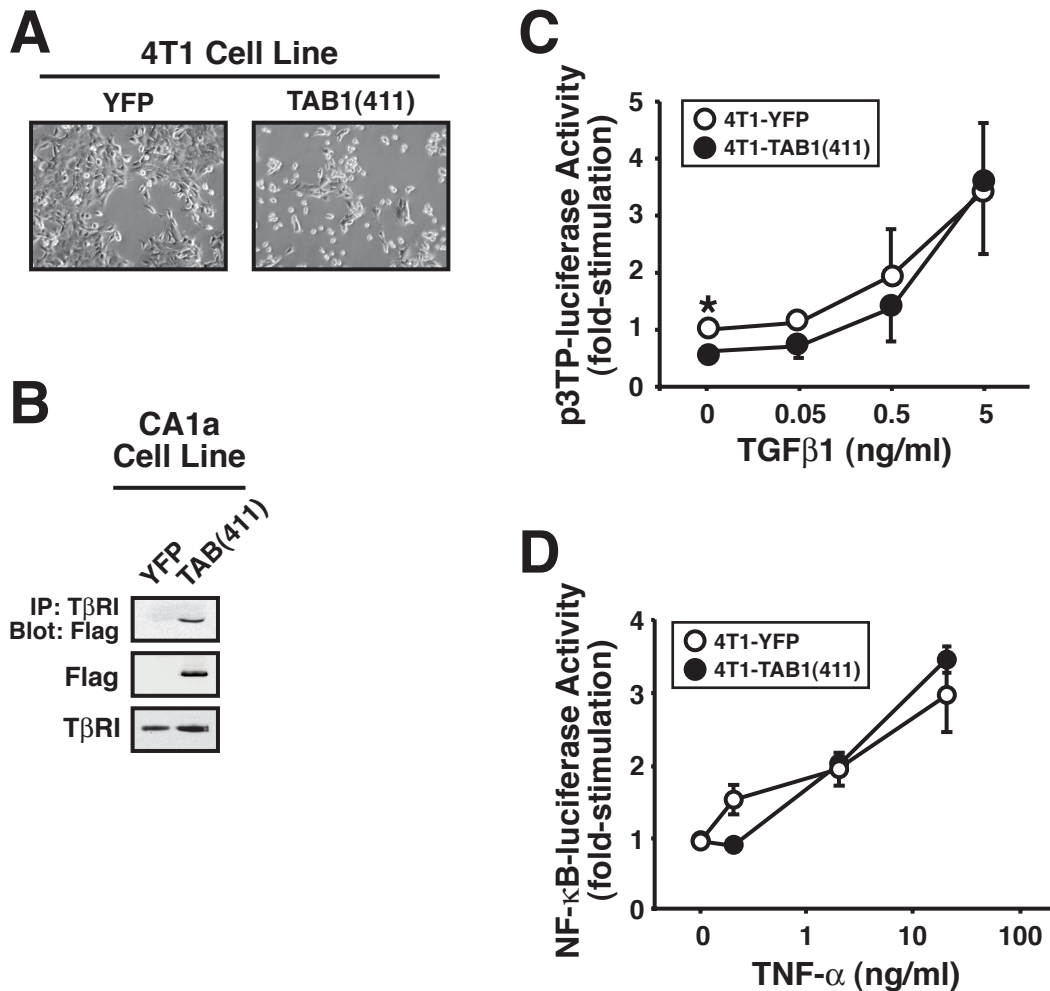


Figure S2. Characteristics of TAB1(411)-expressing breast cancer cells. (A) Phase contrast photomicrographs of control (*i.e.*, YFP) or TAB1(411)-expressing 4T1 cells shows that deficient TAB1 signaling altered the morphology and adhesive characteristics of 4T1 cells. (B) Detergent-solubilized whole cell extracts of control (*i.e.*, YFP) or TAB1(411)-expressing MCF10A-CA1a cells were subjected to immunoprecipitation with anti-TβR-I antibodies. The resulting immunocomplexes were probed with anti-FLAG antibodies to visualize captured TAB1 proteins. Differences in protein loading were monitored by immunoblotting with antibodies for TβR-I and FLAG as indicated. Images are from representative experiment that was repeated once with identical results. (C) Control (*i.e.*, YFP) and TAB1(411)-expressing 4T1 cells were transiently transfected with p3TP-luciferase and β-gal, and subsequently were stimulated with TGF-β1 as indicated. Data are the mean (± SE; n=3) luciferase activities relative to untreated control cells. (*, $P < 0.05$; Student's T-test). (D) Control (*i.e.*, YFP) and TAB1(411)-expressing 4T1 cells were transiently transfected with NF-κB-luciferase and β-gal, and subsequently were stimulated with TNF-α as indicated. Data are the mean (± SE; n=2) luciferase activities relative to untreated control cells. (*, $P < 0.05$; Student's T-test).