

Supplemental Data

Figure S1. Ectopic SAC-GFP induces apoptosis in Par-4-sensitive and Par-4-resistant tumor cells, but not in normal/immortalized cells. Various cancer and immortalized cell lines (from ATCC) were transfected with plasmid expression constructs for pCA-GFP, pCA-SAC-GFP, or pCA-Par-4-GFP, and 24 h apoptotic cells were quantified as described in Materials and Methods. The mouse cell lines are indicated, all other cell lines are of human origin. Note that the SAC domain induces apoptosis in Par-4-sensitive cancer cell lines (PC-3, A549, RM1, H460, CWR22R, LLC1, TRAMP-C2, MCF7/Ras, and HeLa), as well as in Par-4-resistant cancer cell lines (SK-Mel-147, A375-C6, SK-Mel-360, Mel-94, MCF7, and LNCaP), but not in Par-4-resistant immortalized (non-transformed) cell lines (NIH 3T3, BPH-1, PZ-HPV7, MEF, and MCF/10a).

Figure S2. Ectopic SAC-GFP induces regression of non-autochthonous subcutaneous tumors. Tumors were produced in the flanks of nude mice by subcutaneous injections of 1×10^6 cells, and when the tumors had attained a size of 3.5 mm diameter, the tumors were injected with the adenovirus producing GFP, Par-4-GFP, or the SAC-GFP. Tumor sizes were measured over 4-5 weeks and tumor volume was calculated. Note inhibition of tumor growth by the SAC domain in Par-4-sensitive lung cancer A549 and prostate cancer PC-3 derived tumors, as well as in the Par-4-resistant melanoma SK-Mel 147 cell derived tumors. In each experiment, 8 mice were used in each group for adenovirus injection, and data

shown are the mean of three experiments \pm standard deviation bars. The SAC- and the Par-4-adenovirus significantly inhibited tumor growth (* $P < 0.001$).

Figure S3. I κ B-SR inhibits NF- κ B activity in MEFs. Third passage MEFs from littermate control mice were transfected with either NF- κ B-CAT reporter or pGL3-CAT (Empty-CAT) for control, and a β -galactosidase expression construct. These MEFs were subsequently transduced with adenoviral expression constructs for GFP, I κ B-SR, oncogenic Ras-V12, c-Myc, or SAC-GFP. The cell lysates were collected after 16 h, and examined for CAT activity. Relative CAT activity normalized with respect to corresponding β -galactosidase activity is shown.