Supplemental Figure 6. APC generation on MS1 cells following blockade of endogenous TM. a. Blockade of endogenous TM with PPACK-inactivated thrombin and polyclonal anti-TM prevents thrombin-dependent APC generation. Monoclonal anti-TM antibodies, clone 411 and clone 273 (generously provided by Dr. Stephen J. Kennel, University of Tennessee, Knoxville, TN) did not inhibit APC generation. Cells were pre-incubated with one of several antibodies or inactivated thrombin x 1 hr prior to addition of thrombin and protein C. b. Antibody blockade, but not inactivated IIa, had a sustained blocking effect when cells were washed prior to addition of thrombin and protein C. c. Antibody blockade of endogenous TM enabled measurement of dose responsive, fusion protein-dependent protein C activation. Cells were treated with 200nM anti-TM antibody and washed, as above, then incubated with anti-PECAM scFv/TM fusion protein x 30 min, prior to addition of thrombin and protein C.