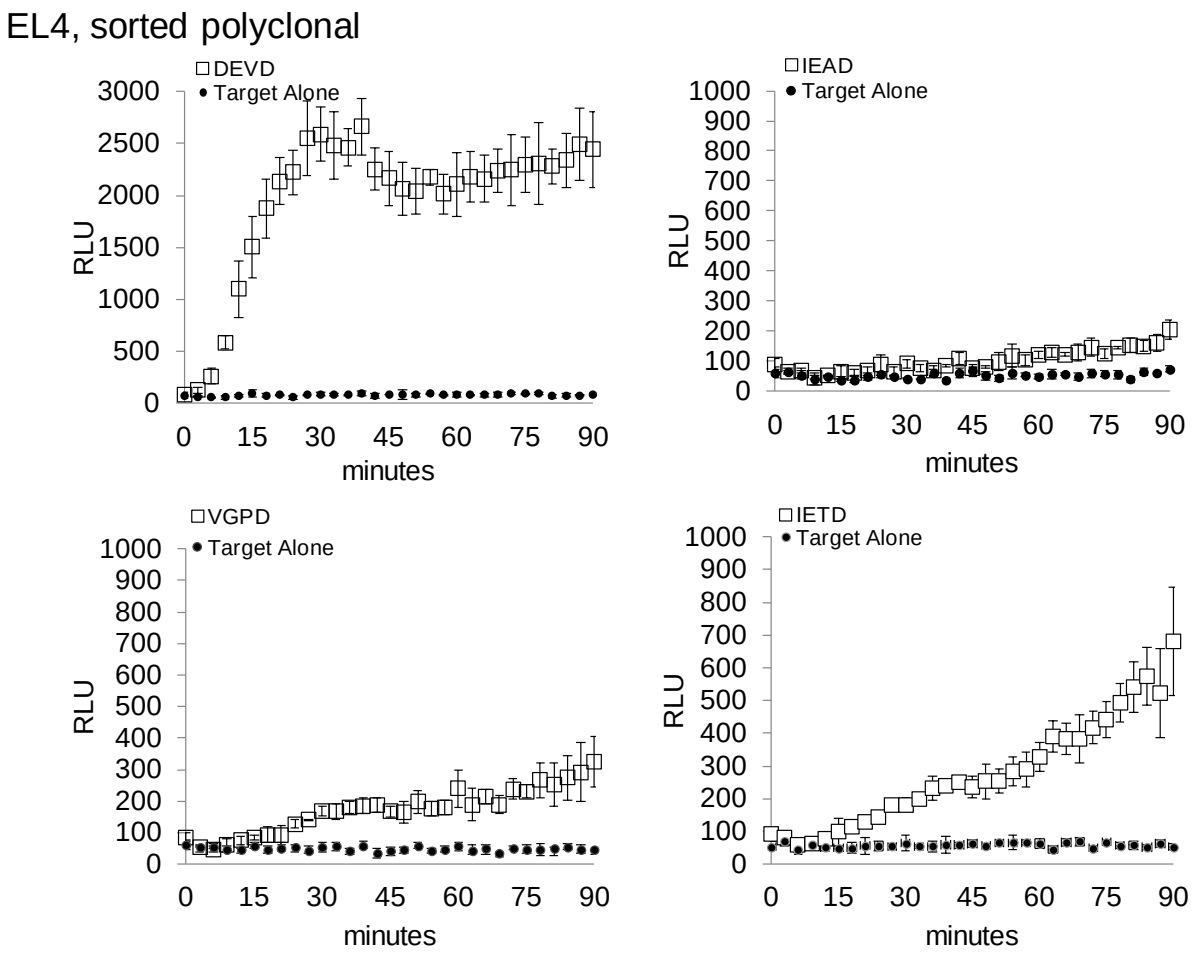
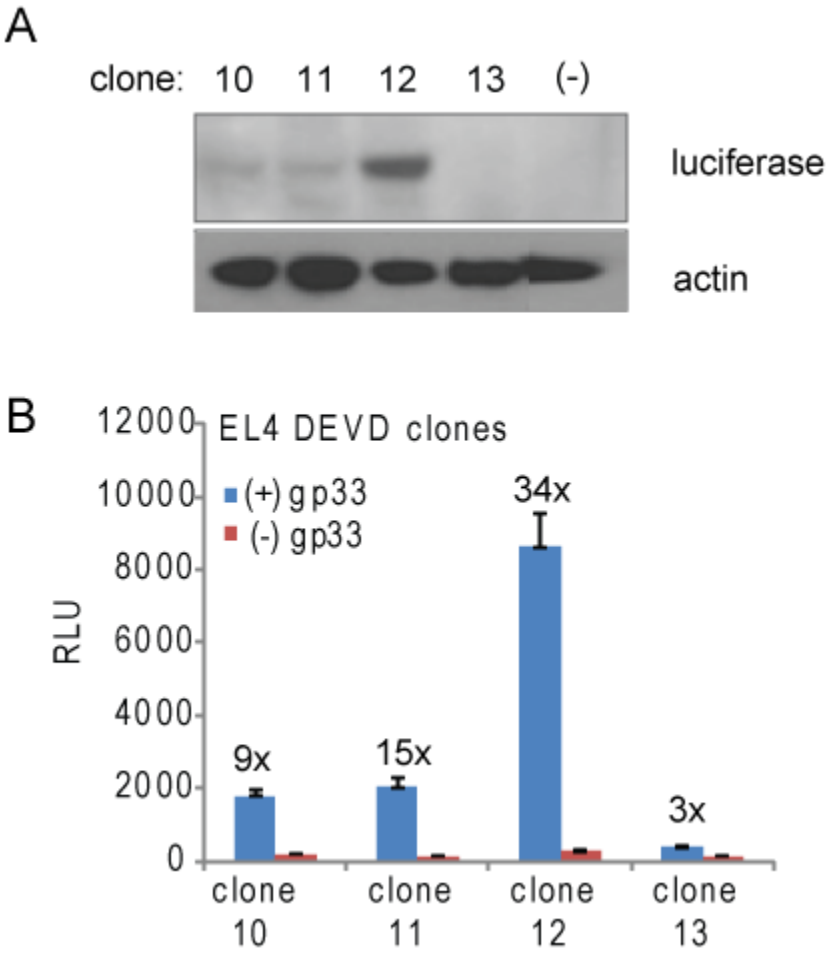
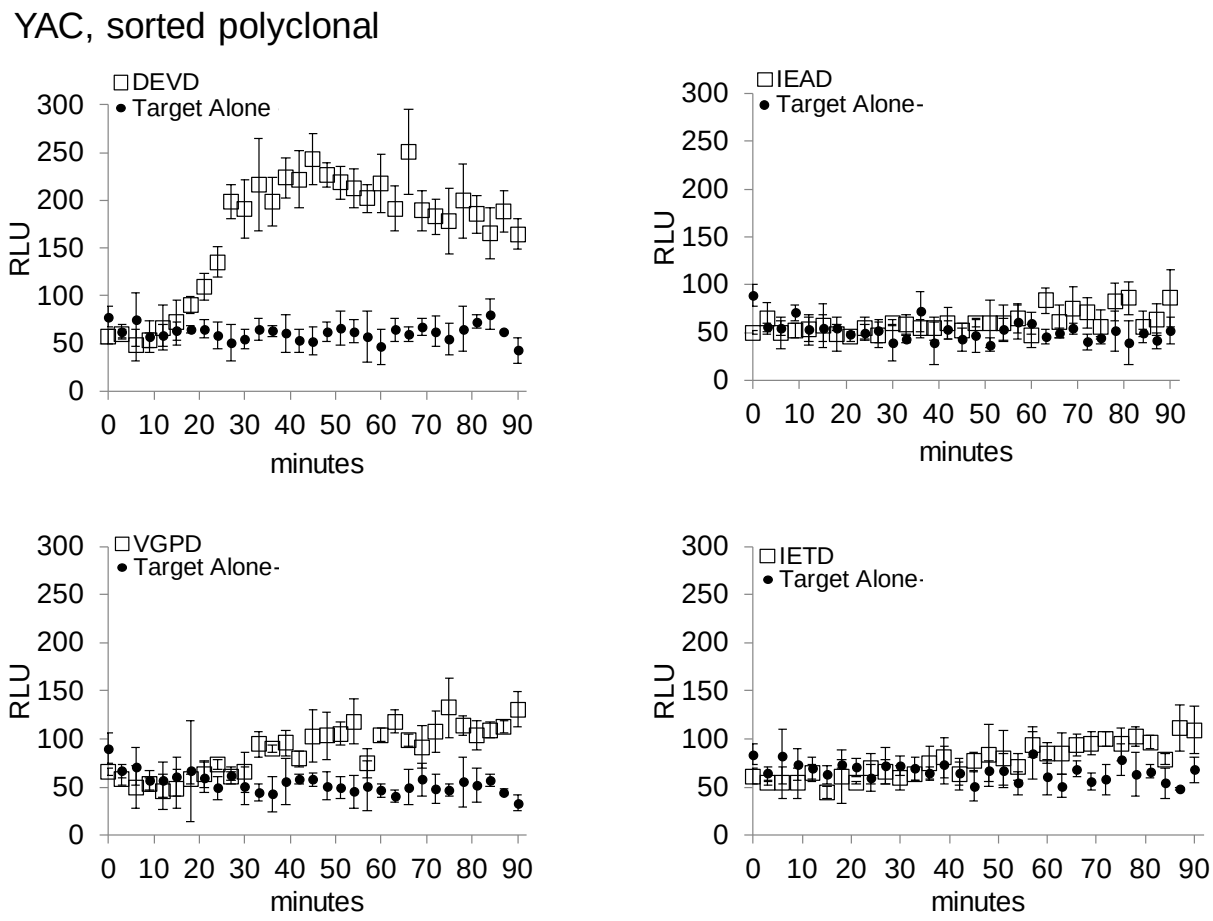


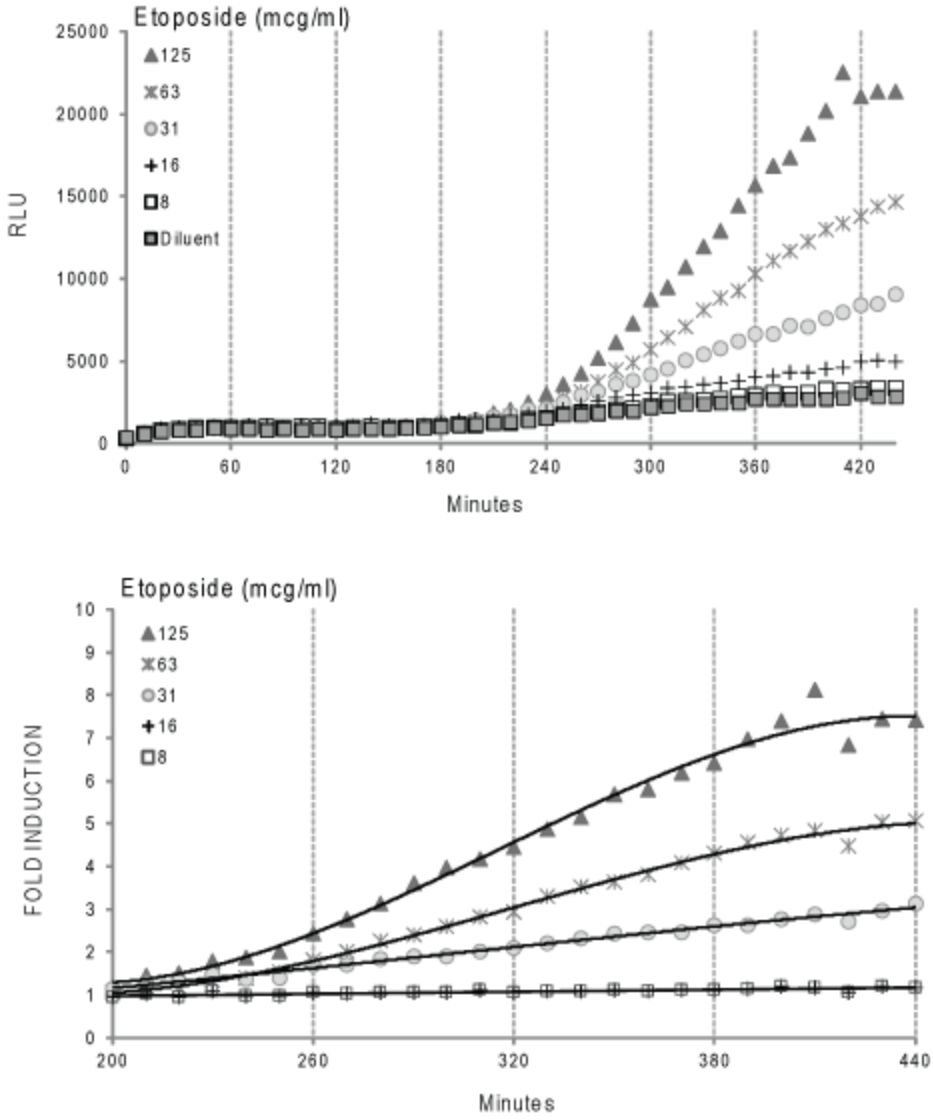
Supplement Figure 1. EL4 cells expressing biosensors. All four biosensor constructs were cloned into a retroviral vector (pMIEG) containing a GFP reporter. EL4 cells were infected with retrovirus and GFP (+) cells sorted at the same mean channel fluorescence. This approach allowed evaluation of a polyclonal, stable population expressing the biosensor. Shown is a co-incubation of luciferin substrate-loaded EL4 cells and P14 CTL at an E/T of 6:1. The baseline luminescence from the biosensor luciferase in the target cells without co-incubation with CTL is indicated by target alone.



Supplement Figure 2. EL4 cell clones expressing GLS.DEVD. (A) Western blot for firefly luciferase in EL4 clones expressing GLS.DEVD. EL4 cells were transfected with the biosensor plasmid and selected by antibiotic resistance. Clones were obtained by limiting dilution; cell lysate was measured for protein expression by western blot with monoclonal anti-firefly luciferase. (B) Induction of GLS.DEVD biosensor by CTL. CTLs were derived by stimulation of splenocytes from TCR transgenic P14 mice with gp33 peptide in vitro. CTLs were co-incubated with substrate-loaded target EL4 clones expressing the GLS.DEVD biosensor at effector/target ratio of 12:1. The target EL4 clones were pulsed with or without gp33 peptide (0.1ug/ml). RLU was measured at 60 minutes. Note that the highest RLU was in clone 12 which expressed the most biosensor protein.



Supplement Figure 3. YAC cells expressing GLS.DEVD. YAC cells are a target for murine NK cells. YAC cells were generated to express either biosensor following retroviral transduction and GFP sort as described above. YAC cells expressing biosensors were then co-incubated with IL-2 stimulated, murine NK cells at an E/T of 1:1. Cytokine activated NK were obtained from C57BL/6 mice spleens using magnetic bead selection (NK cell isolation kit II, Miltenyi Biotec) and maintained in 1000U/ml IL2 for 7 days. No signal above target cells alone was detectable for GLS.IEAD.



Supplemental Figure 4. Induction of caspase 3/7 by etoposide. GLS.DEVD expressing EL4 cells (clone 12) were incubated with titrating concentrations of etoposide in 96 well plates in quadruplicate at 37˚C, and luminescence was measured kinetically every 10 minutes up to 440 minutes as shown in the upper graph. The lower graph shows the fold induction over a restricted time point (200-440 minutes) for comparison to CTL induction of the GLS.DEVD as shown in Figure 4.