



Figure S8. An intact adaptive immune response is necessary for efficacy triple therapy in neu/N mice. (A) Tumor bearing neu/N mice were treated with isotype control (gray line) α CD4 (dashed gray line), α CD8 (dashed black line), or α CD4 + α CD8 (dotted black line) depleting antibodies. Depletion was initiated a week prior to NT2.5 tumor implantation and carried out with biweekly IP injections for the duration of the study. Depletion efficacy of 90% was confirmed before NT2.5 tumor implantation and upon completion of the study. As previously described mice were treated with IT ADU-S100 in sequence with α OX40 receptor agonistic and α PD-L1 antagonistic antibodies. Untreated tumor-bearing neu/N mice were included as control (black line). Tumor

growth was followed for 35 days. **(B)** Serum from tumor-bearing neu/N mice receiving mock or ADU-S100 IT injection sequenced with either α OX40 receptor agonistic and α PD-L1 antagonistic antibodies or isotype matched controls was harvested 7 days post IT injection and HER-2 specific IgG antibody was measured. **(C)** HER-2-specific IgG antibody was measured in the cohorts of mice from figure **(A)** 7 days after IT-ADU-S100 injections. Statistical significance was determined through one-way Anova with significant differences in means re-evaluated using Bonferroni post-test. All data is cumulative of 2 – 3 experiments of 5 mice/group. * $p < 0.05$, ** $p < 0.01$, and *** $p < 0.001$, and **** $p < 0.0001$.